

Preliminary Observations of the Thermal Tolerances of Selected Invertebrate Fauna of Sequim Bay, Washington

The generation of electricity in a thermal plant, either fossil fuelled or nuclear, involves the use of large quantities of cooling water. In coastal areas, the largest source of these cooling waters is the ocean. Although the cooling water effluent may be non-toxic chemically, it will be heated above ambient and in enough quantity to change the temperature of the adjacent water body.

A report on nuclear siting for the Bonneville Power Administration by Battelle-Northwest indicated that potential sites were available on the Straits of Juan de Fuca and within Puget Sound (Harry, *et al* 1967). Although water quality standards for temperatures in coastal water have suggested a permissible elevation of 4°F (2.2°C), many of these are not based on scientific data. The biological effects of thermal addition to the marine coastal environment have not received the same attention as has the freshwater environment and the west coast fauna even less than their eastern counterparts. These experiments were, therefore, conducted to determine the thermal tolerances of various important commercial species.

Thermal tolerance or resistance of the mud snail, *Nassarius obsoletus*, to elevated temperatures has been investigated by Nagabhushanam and Sarojini (1963). They reported tolerances for 1-2 hours of 41°C in sea water and 42°C in air. Fraenkel (1966, 1968) demonstrated the relationship between horizontal intertidal distribution and thermal resistance. Of the 34 species studied, the degree of resistance was found to be closely correlated with the position of the snails in the intertidal zone.

Heat resistance of four intertidal gastropods was investigated by Sandison (1967). A heat coma, characterized by changes in respiration and relaxation of the foot, occurred as much as 10°C below the physiological death temperature of 33-42°C.

Read and Cumming (1967) demonstrated the relationship of thermal tolerance and distribution of three common Atlantic coast bivalves, *Modiolus modiolus* and *Mytilus edulis* were the least temperature-tolerant species and were generally limited in their distribution to the northern latitudes while *Brachidomus demissus*, the most tolerant species, was distributed throughout the tropics.

Additional investigations have been reviewed by Courant (1968, 1969) in literature reviews and in an excellent bibliography by Kennedy and Mihursky (1967); further insight may be obtained through these sources.

The experiments reported here were designed to acquire comparative data regarding tolerance ranges of selected marine invertebrates. As mentioned earlier, a gap exists for specific data that can be readily applied in the marine environment for establishment of water quality standards. These studies were the initial step in filling that gap in the Pacific Northwest.

Methods and Materials

Unfiltered sea water was pumped into the laboratory through 2 inch polyvinylchloride (PVC) pipe into an overhead neoprene-lined tank using centrifugal pumps with polyethylene bodies (Vanton Pump and Equipment Corporation, Hillside, New Jersey). Water was gravity-fed through a 1½ inch PVC pipe into 8 inch diameter PVC pots which contained 4-1200 watt Quartz or Vycor heaters. The water flow from these pots into 100-gallon fiberglass test tanks (Frigid Units, Inc., Toledo, Ohio) was varied from 0.5-1.5 gpm, to deliver the desired temperature range. For very high temperatures, 30° to 40° C, it was necessary to restrict flow and place heaters directly in the tanks; the water was vigorously aerated to ensure even distribution of the water. Polypropylene-lined recirculating pumps (Scientific Supply Co., Seattle, Washington) kept the water mixed. Overflow water was discharged to waste through overflow pipes placed in the tank ends approximately 12 inches from the bottom (Figure 1).

Organisms used in these studies were: the native littleneck clam, *Protothaca staminea*; the Pacific oyster, *Crassostrea gigas*; the Washington or butter clam, *Saxidomus giganteus*; the horse clam, *Tresus nuttallii*; the bay mussel, *Mytilus edulis*; two species of snail, *Thais lamellosa* and *T. lima*; and the Dungeness crab, *Cancer magister*. They were collected at selected points in the northwest corner of Sequim Bay, Washington. The specimens were held in tanks of flowing unfiltered sea water at ambient temperatures (10-12°C) prior to testing.

In the initial studies, organisms were challenged with temperatures between 30°C and 40°C. This proved to be too warm for the local fauna and in later experiments, 30°C was the highest temperature used. Experiments were conducted at increments of 2.5°C±0.5°C in the temperature range between 22.5° and 30°C; i.e. at 22.5° 25°C, 27.5°C and 30°C. The period of exposure during each experiment varied according to the thermal tolerance of the different species. Attempts were made initially to determine the 48-hour TL_m (Median Tolerance Limit) at the different temperatures. However, due to the design of the system and the unpredictable reactions of the organisms, the time necessary for half the individuals to die at a given temperature became the parameter measured.

The determination of mortality varied within the different groups. The criterion of death in the molluscs was not easily applied. Gaping bivalves or snails with their foot extended were prodded with forceps, if the valves did not close or if they did not withdraw their foot, they were counted as mortalities. However, several of the bivalves, most notably oysters and littleneck clams, remained tightly closed. Several of these animals may have been mortalities long before it could be detected, thus a longer time to 50% mortality was indicated than actually occurred. With *Cancer magister*, mortalities were much easier to establish. If no movement could be induced by challenging the crab, or if they could be turned over and no attempt was made on their part to right themselves, they were counted as "mortalities." Many of these "mortalities" recovered in ambient (10°C) sea water; however, it was assumed they would have perished if left at the elevated temperatures.

Results

The early studies indicated that the organisms could not tolerate 35° to 40°C. The time to death at these temperatures among all specimens used was measured in minutes or seconds.

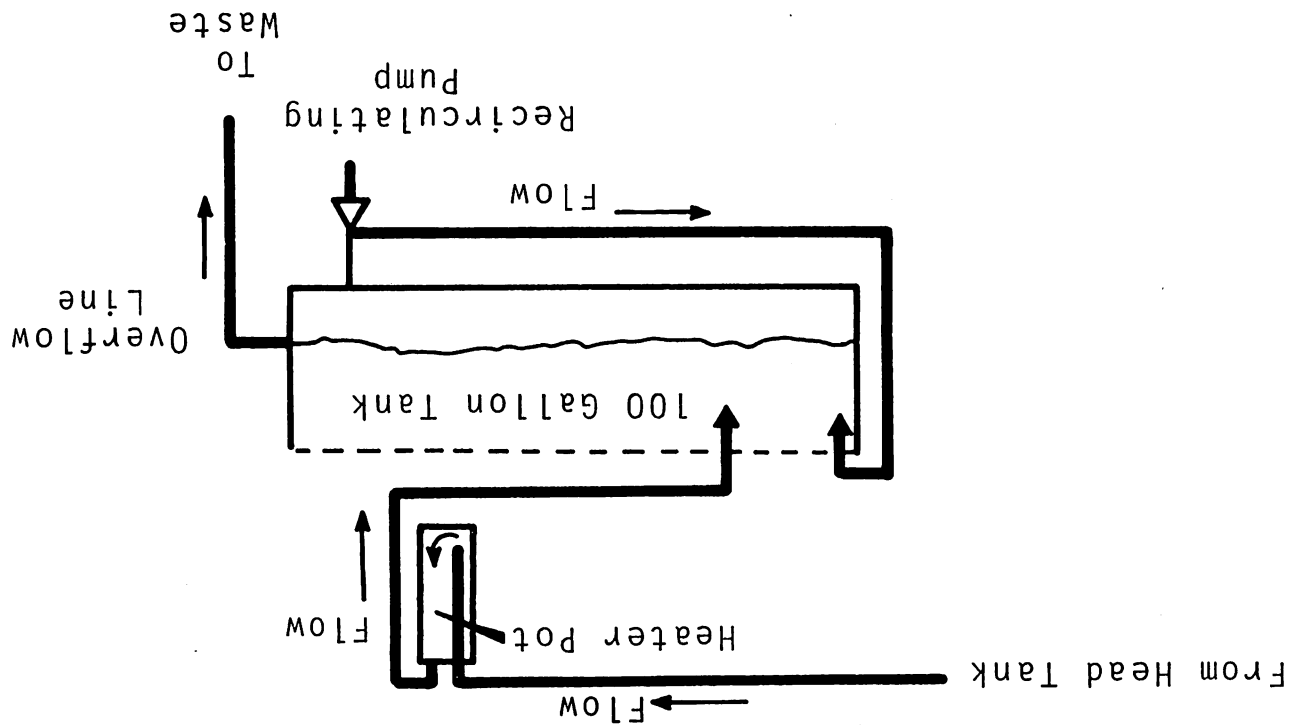


Figure 1. Diagrammatic representation of system used in thermal experiment (no scale).

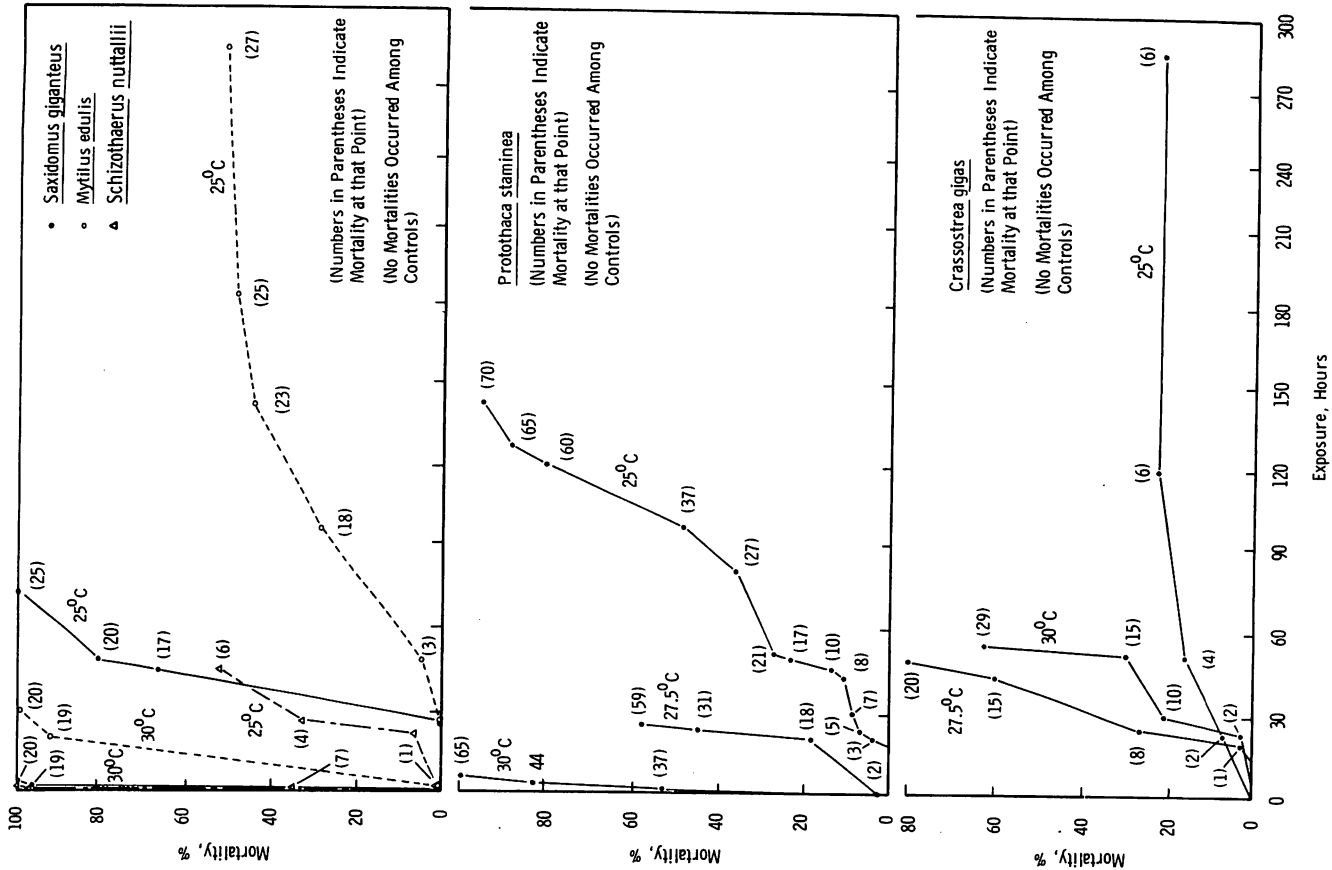


Figure 2. Pelecypod mortality with the passage of time at different temperatures.

Figure 2 indicates the time for 50% of the organisms to die at a fixed constant temperature. Although there is an obvious need for additional information several observations are considered significant. The most sensitive species were the butter clam, *Saxidomus gigas*, and the horse clam, *Tresus nuttalli*. Bay mussels, *Mytilus edulis*, and Pacific oysters, *Crassostrea gigas*, were the most resistant species; the native littleneck clams, *Protothaca staminea*, were intermediate between butter and horse clams and the mussels and oysters.

The graphed results for *C. gigas* at 27.5°C and 30°C appear to be reversed, i.e. it is indeed odd to have a higher mortality rate at the lower temperature. A need for replication with a larger sample size is obvious.

Figure 3 shows the time until death of two closely related, both specifically and ecologically, species of snail: *Thais lamellosa* and *T. lima*. No real difference could be determined; the curves nearly overlay one another. Replication with larger sample sizes may elucidate minor differences.

Controls elicited no mortality during the experimental periods.

Crab

All *C. magister* used in these studies were less than 80 mm across the carapace.

At 30°C, 100% of the *C. magister* were motionless and appeared dead within 2 minutes, apparently victims of thermal shock. However, when returned to ambient temperatures (12°C), all specimens were able to recover and apparently return to normal activities.

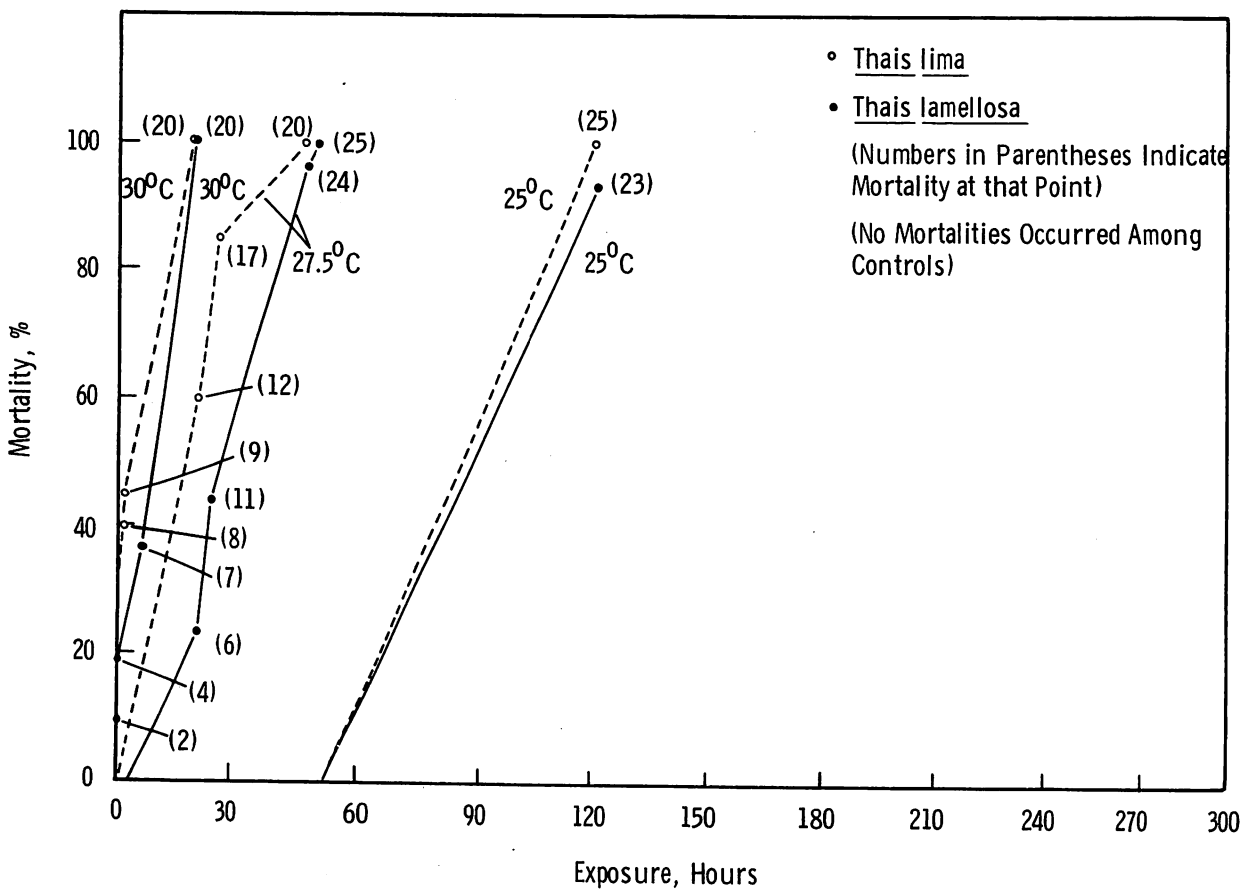
When placed in 27.5°C, the Dungeness crabs immediately experienced the same type of shock. There was an initial loss of equilibrium followed by an apparent extreme irritation during which they continually brushed their mandibles with their chelipeds. Following this period of excitation, the animals were relatively calm, but the mandibles continued to move rapidly. Behavioral changes also occurred; the specimens became very docile and would not defend themselves when challenged. Within 18-24 hours, 100% mortality occurred.

At 25°C, the crabs had essentially the same reaction. There were the early symptoms of shock; recovery was rapid, usually within 60 seconds. Activity increased until they seemed normal; no mortalities occurred within 168 hours.

In the only attempt to increase resistance by acclimation, 5 crabs were placed into 22.5°C water for 72 hours and then transferred into 27.5°C water. There was no apparent initial shock, nor any decrease in activity or mortality in the next 48 hours. However, one crab, which attempted to molt, died. No mortalities occurred among the controls.

Discussion

Adams (1969) pointed out that many laboratory experiments have been conducted on marine aquatic organisms to predict the effects of warm water effluent which in no way resemble the conditions experienced at operating thermal plants. For example, he feels it is useless to subject an organism to a 24 or 96 hour temperature tolerance test when the exposure of an organism to a temperature elevation at a particular site is often a matter of minutes rather than of hours or days. In these experiments,



however, it was deemed necessary to attempt to define a 48 hour TL_m for the purpose of being able to compare these results with those in other laboratories involving the same type of organisms. For example, Dickie (1958) studied the scallop, *Placopecten magellanicus*, and found the 48 hour limit (for specimens acclimated at 10.5°C) to be 23°C. This compares with the less resistant species examined in these studies. Further, determining resistance time-temperature relationship is an indicator of the greatest temperature these sessile animals can tolerate and for how long. Such data could be useful in predicting the outer limits of distributions of these organisms given a physical description of temperature patterns around a heated water diffuser.

Molluscs

As indicated, death within this group was often difficult to establish. If it was not possible to demonstrate lack of tactile response or if the response was weak, the organisms were not listed as mortalities. Dickie (1958) however, pointed out the need to include "weak" animals in the mortality category; with scallops, weak animals invariably died when replaced in ambient temperature sea water.

The data indicate a relationship between intertidal height of animal occurrence and thermal resistance. *Saxidomus giganteus* and *Tressus nuttallii*, killed within 72 hours at 25°C, are found much deeper in the clam beds than *Protothaca staminea* (dead after 144 hours at 25°C), while both *Crassostrea gigas* and *Mytilus edulis* rest on the surface of the beach and were relatively healthy after 12 days at 25°C. Therefore, as one moves vertically across the intertidal zone toward the water, the three most resistant species are also found higher in the intertidal zone.

The similarity in resistance of the two *Thais* species is not surprising. Both occupy essentially the same intertidal level. When adapting to such a level, both species apparently developed many of the same tolerances; generally they are found intermixed in their particular intertidal zone.

Crab

The young Dungeness crab must be well adapted to a wide range of temperature during their first year. They spend a certain amount of their early benthic life in the shallows during the summer months where water temperatures may rise rapidly during a midday low tide. These crabs must be adapted to a wide temperature range to ensure species survival until such time as they move to somewhat deeper water. No studies, however, were conducted with adults to determine whether they maintain or lose their resistance.

A higher ambient temperature (15-20°C) or the use of an intermediate acclimation temperature (20-22.5°C) may have produced markedly different results. The process of acclimation, i.e. holding the organisms at some sublethal but higher temperature, may have given the experimental organisms the ability to withstand the temperatures which proved to be fatal. Therefore, a true lethal temperature would depend on the rate and actual temperature of acclimation.

The acclimation of 5 crabs at 22.5°C indicated the marked ability of these crabs to adapt. Jones (1964) pointed out that for any particular acclimation temperature, every organism has a temperature range within which "existence" for an indefinite period is possible. This range has an upper limit above which the animal cannot live

indefinitely but survives for some limited period. Below the thermal death-point the organisms demonstrate "tolerance"; at temperatures above the upper limit, the organisms demonstrate "resistance." Fry (1947) defines the thermal death-point as that temperature above which 50% of the population cannot live indefinitely. Below this is the "zone of thermal tolerance" and above it, the "zone of thermal resistance." Above the thermal death-point, the resistance time shortens with a progressive rise in temperature until a point is reached at which the animal is killed instantaneously when transferred from water at the acclimation temperature to water at test temperature. The rise in the thermal death-point is less than the rise in the acclimation temperature.

Although these crabs did survive with acclimation at a temperature which proved fatal without acclimation, problems in growth at 27.5°C may have been demonstrated by the molting mortality. Additional work in the area of sublethal effects from continued exposure to constant temperature as well as fluctuating temperatures is obviously needed.

Although the temperatures reported here were lower than could cause acute mortalities in the immediate vicinity of a heated water diffuser, these studies were deemed necessary to establish the range in which further investigations would be carried out. Adams (1969) has shown heated discharge waters at the Pittsburg, California Power Plant to drop from an increase of 10°C to 1-5°C within 14 minutes. The studies reported here establish the relationship between these and other laboratory investigations. Further experiments will not exceed temperatures expected in the near vicinity of a heated water diffuser, nor will they maintain organisms at those temperatures for longer than the expected time period; i.e. studies will examine increases of 10°C for 30 minutes or less, 8° for longer periods, etc.

Summary

1. A study was conducted to examine the thermal tolerance of seven species of molluscs and the Dungeness crab, *Cancer magister*.
2. Among the pelecypod molluscs, a relationship was observed between height in the intertidal zone and tolerance to elevated temperature. *Crassostrea gigas*, the Pacific oyster, and *Mytilus edulis*, the bay mussel, are found above ground and at higher tidal levels; these organisms were the most resistant. On the other hand, *Saxidomus giganteus*, the butter clam, and *Tressus nuttallii*, the horse clam, are found buried in the substrate (and at lower tidal levels) and were the least resistant.
3. *Cancer magister* was demonstrated to tolerate elevated temperatures of $\Delta 15^\circ\text{C}$. Following acclimation, this was increased; however there is an indication growth may be inhibited.

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The Muskegs of Southeast Alaska and Their Diminished Extent

Southeast Alaska receives abundant, year-round precipitation and has cool summers. Precipitation exceeds calculated evapotranspiration throughout the year in most of the area, and three-fourths of the stations have projected moisture deficits of less than 0.1 inch (Patric and Black, 1868). Mainly because of the late Pleistocene glaciation, it also has many depressions and extensive impermeable soil layers (commonly compact glacial till or waterlaid glacial flour). As a result of these factors, there are extensive areas of organic soils.

The term "muskeg" according to Dachnowski-Stokes (1941) denotes "an area covered with sphagnum mosses and tufts of sedges." In southeast Alaska, all relatively open peat bogs that have a ground cover high in sphagnum mosses and/or sedges are called "muskegs."

The muskegs of southeast Alaska vary in depth, composition, and vegetative relations. Depth of organic soil material ranges from less than two feet to over 40 feet. They may be composed of sphagnum peat, sedge peat, or muck, usually with some component of wood. Vegetation growing on a muskeg is related to type of organic material, which in turn is related to water table regime and movement. In addition, a large acreage of wet organic soils supports forest vegetation.

A few publications have discussed the muskegs of southeast Alaska with conflicting interpretations of whether forest or muskeg is expanding. Zach (1950) and Lawrence (1958) have postulated that muskeg, except on steep slopes, is the climax vegetative type in southeast Alaska. They point out that most open peat soils contain wood at some depth. They also interpret the presence of spike-topped and dead trees near the margin of muskegs as evidence that muskegs are enlarging.

Dachnowski-Stokes (1941, p. 32) and Heusser (1960, p. 187) both state (without quantitative evidence) that forest is invading muskeg in southeast Alaska.

New evidence, developed from soil surveys and other detailed soil investigations, shows that muskeg area has diminished greatly in the recent geologic past.

Methods

The Alaska Region of the U.S. Forest Service has been conducting soil surveys in southeast Alaska since 1961 when the Hollis Area soil survey was initiated. This survey has been completed (Gass, *et al.*, 1967) and other surveys are now underway or completed (Fig. 1). As National Cooperative Soil Surveys, the soils are classified