Population Ecology of the Sonoran Mud Turtle (*Kinosternon sonoriense*)

at Quitobaquito Springs, Organ Pipe Cactus National Monument, Arizona

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FINAL REPORT

on the funded project

Status and Conservation of the Río Sonoyta Mud Turtle
Kinosternon sonoriense longifemorale at Quitobaquito Springs

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Executive Summary

This report covers 14 years of study of the Sonoran mud turtle at Quitobaquito Springs, Organ Pipe Cactus National Monument, Pima County, Arizona, 1982-1995. The population represents a subspecies endemic to Quitobaquito and nearby Río Sonoyta in Mexico, the Río Sonoyta mud turtle, *Kinosternon sonoriense longifemorale*. The Río Sonoyta population cannot be assumed secure due to agricultural and urban development. Previous research indicated that the Quitobaquito population had declined. The Quitobaquito turtle population lives in the primary refuge for the desert pupfish, an endangered species.

Results are presented in this report on the following characteristics of the Sonoran mud turtle population: age-specific habitat use, population size and structure, age-specific and sex-specific survivorship, reproduction and growth rates, sex ratio, recruitment and population trends, mortality, habitat chemical composition, pesticide contamination, lipid storage, and food availability. The findings permit construction of a life table for this population--the first available for this species. The life table assists integration of the natural history and ecology of the population to reach a synthesis on population processes and trends. This provides a strong basis for understanding the population, and for guiding continued study, monitoring, and management.

Sampling was conducted for a total of 74 days of trapping totalling 1099 trap-days with hoop nets plus 470 trap-days for modified minnow traps used for hatchlings and juveniles. Additional records were obtained by hand capture, visual observation, reports from other field personnel, and radio-telemetry, for a total of 560 turtle records, including 273 recaptures of marked turtles.

Annual adult female survivorship at Quitobaquito was estimated at 85.5%, somewhat below most previous reports for this species. Male survivorship was higher at about 90%, and the sex ratio is nearly 2:1 in favor of males, with the survivorship difference possibly explaining most of the imbalance. Juvenile survivorship is lower, increasing gradually to adult levels. Egg survivorship appears to be high, and was estimated at 85-90%; eggs of this species are known to hatch about a year after laying, and have embryonic diapause. Sex determination depends upon incubation temperatures at a critical period (it is likely that females are produced by warm temperatures in the spring prior to hatching (M. Ewert, personal communication). This might also contribute to the sex ratio anomaly. Clutch size averaged 4.0 eggs (n=3). Clutch frequency was estimated at 1.5 clutches/year/female, which is rather low for this species. Females first produce a clutch at just under 6 years of age. Males mature at age 4 years, probably as a function of size rather than age. Growth is moderate in early life, and slows abruptly at maturity.

A substantial number of mud turtle carcasses (n=22) were recovered at Quitobaquito during the study period, many of which displayed no sign of injury. Analysis of water and sediment quality at Quitobaquito, and of chemical composition of 8 of the turtle carcasses that were recovered relatively fresh, yielded no convincing evidence sustaining an ecotoxicological hypothesis involving organochlorine pesticides or heavy metals. Low levels of DDT metabolite were confirmed in 4 of the carcasses, and certain elemental metals may have been present at elevated levels. Water and sediment had modestly elevated arsenic levels, but the carcasses did

not contain arsenic concentrations likely to be toxic. We should remain vigilant about possible contamination or natural toxicity at Quitobaquito nonetheless. The carcasses all had remarkably low levels of stored fat, and it appears that the turtles are undernourished, and are feeding heavily on plants, a non-preferred food.

Aquatic invertebrate availability as food for mud turtles at Quitobaquito was apparently in short supply. There is good evidence that the great success of the desert pupfish at Quitobaquito results in depressed food availability for the turtles. If so, the unusual mortality observed in turtles is a primarily natural phenomenon resulting from competition with a native species population that faces very few effective predators. Competition between fish and turtles has not previously been reported, although it is probably both frequent and important.

The population at Quitobaquito probably numbered in the hundreds in the late 1950's, and was buffeted by early heavy-handed early management efforts directed at saving the pupfish. By 1970 it was clear to park naturalist Scotty Steenbergh that the population had declined substantially, and Dr. Fred Gehlbach estimated the population at 143 individuals (hatchlings not included in the estimates). Early in the 1980's, we estimated about 100 individuals, with a continuing decline into the late 1980's. We earlier suggested that low survivorship among juvenile turtles, as a result of degradation of shallow-water, well-vegetated habitat for juveniles in early management actions, was primarily to blame for the population decline. The additional data and analysis here supports this hypothesis. In winter 1989-1990, the Park Service recreated a substantial amount of shallow, well-vegetated habitat for juvenile mud turtles, along the lines recommended by us and other herpetologists. Early results for the 1990s show increased juvenile survivorship, and population estimates significantly above the late-1980's minimum, at about 130 individuals.

A life table was constructed for the Sonoran mud turtle population at Quitobaquito utilizing the best available estimates for all of the relevant parameters (survivorship, clutch size and frequency, age at maturity, sex ratio at hatching). The result yielded a generation time of 12 years, and net replacement rate $R_0 = 1.6$, indicating a population growing at the rate of 60% per generation. Sensitivity analysis of the life table, employing parameter variation in the ranges indicated by the uncertainty of the various estimates tended to confirm a stable, and probably growing, population. The strongest negative effect on R_0 was caused by varying juvenile survivorship to mimic conditions observed in the mid-1980's. Early results suggest that the population may be recovering in response to management efforts.

Recommendations are for (1) research and monitoring to confirm or revise the observed population trends and life history trait estimates, (2) more detailed study of nesting in another desert population of the Sonoran mud turtle, (3) vigilance against the appearance of vertebrate mortality or chemical toxicity at Quitobaquito, and (4) international collaboration in a study of the Río Sonoyta mud turtle in Río Sonoyta, Sonora.

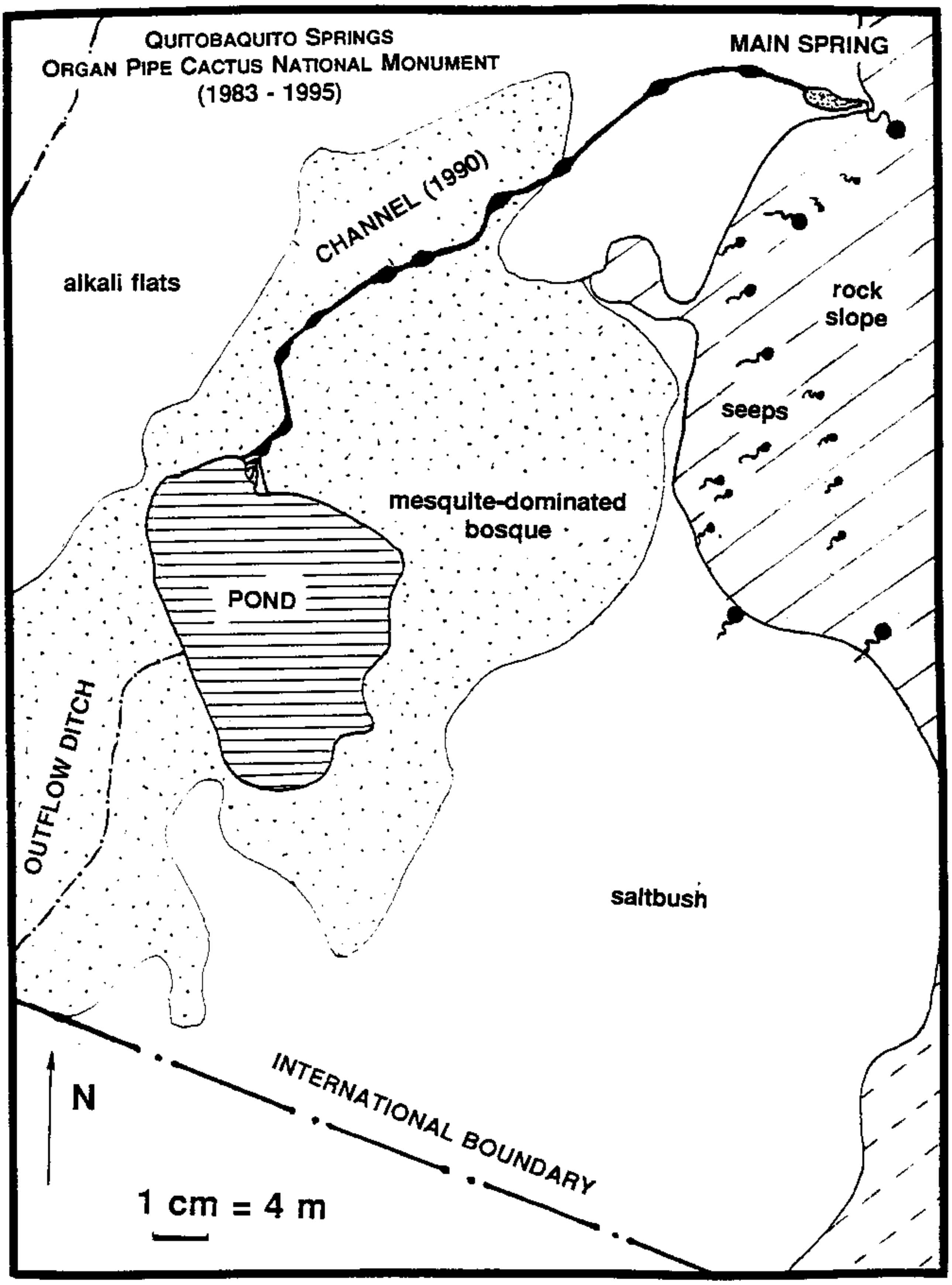


Figure 1. Semi-diagrammatic map of the study area.

INTRODUCTION AND HISTORY

The Sonoran mud turtle (*Kinosternon sonoriense*) occurs at Quitobaquito at an arid western margin of its geographic distribution. There, and at Río Sonoyta within a few miles of Quitobaquito, a hydrologically isolated deme of the species persists as a described subspecies, the Río Sonoyta mud turtle (*K. s. longifemorale*; Iverson, 1976, 1981). At Quitobaquito, on the Mexican border in Organ Pipe Cactus National Monument (ORPI), the mud turtle lives in a man-made and maintained pond, spring pool, and channel habitat (Fig. 1) that is the main refuge for the once widespread but now Federally Endangered desert pupfish (*Cyprinodon macularius*; Kynard, 1976; Miller and Fuiman, 1987).

The Sonoran mud turtle is a strongly aquatic southwestern species that occurs in perennial to semi-perennial waters (Hulse, 1974a; Rosen, 1987). It lives in a remarkable variety of waters ranging from tiny springs and tinajas to the mainstream in such large rivers as the Salt and Verde in Arizona (personal observations). It is an omnivore that prefers invertebrates and fish when available (Hulse, 1974b), although it is inefficient at catching fish (personal observation). The life history is variable, reflecting the great habitat breadth (Rosen, 1987; Hulse, 1974a&b, 1976a&b, 1982; Van Loben Sels et al., 1995).

Quitobaquito is an oasis associated with a series of natural springs rising in fractured granites along the southwest faces of the Quitobaquito Hills in the low, arid southwestern portion of ORPI. The springs were well known and regularly used by early caucasian travelers and colonizers (Greene, 1977; Bennett and Kunzmann, 1989; e.g., Hornaday, 1908), as well as by indigenous peoples during (Nabham, 1982; Bennet and Kunzmann, 1989) and undoubtedly prior to caucasian arrival. Quitobaquito was a major way station (Agua Dulce, on Río Sonoyta a few miles west, being the most westerly) for the passage across the dangerously hot and dry Camino del Diablo to the Colorado River. It was inhabited, farmed (via irrigation), and in caucasian times grazed by domestic livestock, for hundreds, and possibly thousands, of years.

Since 1957, when the National Park Service (NPS) purchased the area from Jose Juan Orosco for the then-lofty sum of $14,000, the area has been under full NPS authority, dedicated to non-consumptive purposes including picnicking, bird-watching, pupfish conservation, and more recently, research on and conservation of turtles, snails, and specialized plants (Bennett et al., 1990, Brown and Warren, 1986; Cole and Whiteside, 1965; Felger et al., 1992; Fisher, 1989; Johnson et al., 1983; Kingsley and Bailowitz, 1987; Kingsley et al., 1987; Landye, 1981; Petryszyn and Cockrum, 1990; Warren and Anderson, 1987).

With the cessation of grazing and irrigation ditch maintenance in 1957, Quitobaquito quickly developed a choking growth of emergent vegetation, primarily tule (or American bulrush, *Scirpes olneyi*--more recently termed *S. americanus*), that threatened to close all open water in the pond and block the spring flow maintained via pipes tapped back into the source (Bennett and Kunzmann, 1989). It would appear that the natural vegetation climax at the site is a sedge meadow with little or no open water, and thus insufficient habitat for endangered fish, or for that matter, turtles.

As of 1983, desert pupfish still occurred in Río Sonoyta (although these are not identical with the Quitobaquito pupfish, *C. m. eremus*) as a rare species, along with native longfin dace (*Agosia chrysogaster*), and introduced black bullheads (*Ameiurus melas*) and mosquitofish (*Gambusia affinis*). Exotic fishes in the desert southwest, including those in Río Sonoyta, are primarily blamed for the severe decline of most of the regional fish species (Minckley and Deacon, 1991). In addition, the aquifer in Río Sonoyta Valley has been developed from the 1970's as the source for a major pumping-based irrigation agriculture center, seriously threatening stream flow and thereby threatening the pupfish and mud turtle. Since the river cannot be relied upon, Quitobaquito must be kept and physically maintained as a refuge, foremost for the pupfish, and within that framework for the mud turtle and other unique or unusual biota.

Upon discovery of the hydrologic problem developing at Quitobaquito Pond in the late 1950's, Lowe and fellow herpetologist Dr. Howard K. Gloyd recommended that NPS physically maintain spring flow, and further, that the pond be deepened. They recommended that a primarily shallow pond be created. In the event, the pond was dried and bulldozed in 1961 into a too-deep, straight-walled pool with a small shelf of shallow water. As detailed herein, this shallow water is critical to the turtle population, and its small size is a factor in the mud turtle population decline. Spring water was later re-routed through an underground pipe, eliminating ditch habitat for a rare snail, and again, eliminating shallow water habitat for juvenile turtles. The pond was dried out again in 1969-1970 to remove introduced shiners that were a threat to the pupfish. At that time, Warren (Scotty) Steenbergh, park naturalist, already noted that mud turtles had declined since the first NPS management effort.

During both NPS pond-draining episodes, turtles were, at best, ignored, and many were apparently given away as pets (Rosen, 1986; Bennett and Kunzmann, 1989). Since earlier decades, and since 1971, the population of Sonoran mud turtles at Quitobaquito appears to have declined significantly, and population densities in the 1980's were well below those expected based on the species' norm (Rosen 1986, 1987). The population at that time appeared to remain in decline (Rosen and Lowe, 1996 *in press*). Based on Rosen's (1986) recommendation that juvenile turtle habitat should be restored, NPS in winter 1989-1990 created a semi-natural spring channel, modelled after Tule Creek, Yavapai County, Arizona (Rosen, 1986, 1987), delivering water through a 100+ meter course from main spring to pond. At the same time, a small shallow area in the northeast pond corner was excavated from the tule mat; and based on the odd suggestion that mud turtles, which are excellent climbers, could not climb out of the pond to nest (Bennett and Kunzmann, 1989), a "nesting island" was installed near these shallows.

Herpetologists have visited Quitobaquito from their earliest presence in the region (Lowe, 1987), and have continued to visit and study turtles there regularly and frequently (Smith and Hensley, 1957; Hulse, 1974a, 1976a&b; Gehlbach, 1979; Iverson, 1981, 1989; Rosen, 1986, 1987). Smith and Hensley (1957) collected and preserved a mating pair of yellow mud turtles (*K. flavescens*) at Quitobaquito in 1955, the only record of this species for the region, and possibly reflecting an introduction of turtles picked up on Ajo Way between Tucson and ORPI. Hulse (1974a) removed an introduced painted turtle (*Chrysemys picta dorsalis*), Bennett and Kunzmann obtained an exotic cooter (*Pseudemys* sp.), and we removed a red-eared slider (*Trachmeys scripta*) as well as an adult bullhead catfish during the 1993 work described herein. Many herpetologists have trapped turtles at Quitobaquito, without doubt more than we know. The number of turtles scientifically collected from the site is fairly substantial, although the known number in no way suggests an effect on population size. Visitors, and in an earlier epoch NPS personnel, are well known to have removed mud turtles, probably in far greater numbers than done by the scientific collectors.

The present study was initiated in 1982-1983 by NPS personnel, Peter Bennett and Michael Kunzmann, at the University of Arizona Cooperative National Parks Service Study Unit (CPSU). In 1983-1985, the work was contracted to Rosen during Masters Degree work at Arizona State University, although Bennett, Kunzmann, Lowe, and Lowe's student David Hall conducted some sporadic, but very limited, operations well into early 1985. All available data from this early work has finally been turned over to Rosen, and an early report of immigration of a turtle from Río Sonoyta to Quitobaquito (Rosen, 1986) is now known to be an accidental release of a marked turtle at Quitobaquito (Rosen, personal observation; Peter Holm, personal communication). In any event, Rosen re-censused the population in fall 1989 in anticipation of NPS habitat improvement construction during the coming winter (above). Based on recommendations by Lowe and Rosen (1992; see Rosen and Lowe, 1996 *in press*), ORPI maintained basic visual monitoring of turtles and collection of turtle carcasses, for post-mortem analysis, 1990-present. In fall 1992 in anticipation of the larger effort in 1993 made possible by Arizona Game and Fish Department (AGFD) Heritage Grant funding to Rosen and Lowe, we carried out an intensive trapping census, followed in July-August 1993 by a more extended trapping episode. In September 1995, following as yet-unverified reports of more exotic fishes at Quitobaquito, ORPI Resources Management personnel Charles Conner (trained by Rosen in turtle study at Quitobaquito) and Tim Tibbitts conducted a successful turtle (but exotic fish-free) trapping session at the pond, forwarding the data for this report.

This report details relevant life history and ecological findings related to the low population density and apparent population decline of the Sonoran mud turtle (*Kinosternon sonoriense longifemorale*) during the 1970's to 1990's at Quitobaquito. In addition to low juvenile survivorship, a problem now possibly solved, the report discusses the vexing appearance of inexplicably dead turtles and the possibility that resource stress or contaminants may be to blame. A brief foray into radio-telemetry study is described, and the early returns on the effects of the NPS 1989-1990 turtle-habitat management efforts are evaluated.

METHODOLOGY

The primary methodology was mark-release-recapture based on repeated sampling using baited turtle traps, as detailed by Rosen (1986, 1987). Two principal trapping methods were used. Adults and larger juveniles were caught in hoop nets with 2.54 cm (1") mesh and single 30.5 cm (12") funnel entrances. These were usually baited with two oil-packed sardines plus one hotdog, although fresh fish, dogfood, and tunafish were also tried occasionally. We usually set approximately 20 of these traps. Hatchlings and small juveniles were caught in commercial minnow traps made of 0.64 cm (1/4") wire mesh with both funnels opened to 7.6 cm (3"); we usually set about 15 of these traps, each baited with half of a sardine.

All traps were checked daily, and bait was replaced after two days or whenever it disappeared or became rotten. Traps were set along banks, facing root masses, in weed beds, and in productive areas of deep water. Whenever possible turtles were caught by hand or dipnet, but this resulted in a minor proportion of captures. Minnow traps captured many more fish and invertebrates than turtles, and complete records were kept of these.

All captured individuals were measured, weighed, and individually marked by notching marginal scutes with a hacksaw, scissors, or clippers. Marks were made and recorded by counting from the first marginal (posterior to the nuchal) back, on anatomical left or right. For example, 1R,2R,11L had the first and second marginals notched on the right side, and the last (11th) on the left side. Early in the study, bridge scutes (marginals 5-7) were notched, but this was discontinued in mid-1983. The following data were taken: carapace length (CL in mm, straight line parallel to main axis of animal); plastron length (PL in mm, exactly on midventral line); total weight in grams; presence of active growth zones (annuli) on plastral scutes; measurements of visible annuli on the most clearly readable plastral scute (to $\pm$ 0.1 mm; usually on the abdominal); reproductive status; and location. Maximum shell width and height data were also recorded for most captures. All females $\geq$ 80 mm CL (well below the minimum size of maturity for this species) were checked for oviducal eggs by palpation, and a few were taken to an X-ray facility for determination of clutch parameters (Gibbons and Greene, 1979) and released shortly thereafter.

A procedure for estimating clutch frequency was developed using selected dissections in two other populations, Sycamore Creek at Sunflower (Maricopa County) and Tule Creek (Yavapai County), Arizona (Rosen, 1987), combined with palpation and X-ray results from Sonoran mud turtle populations at several sites across Arizona. This more extensive data set allows a reasonable approximation of annual clutch frequency at Quitobaquito, where reproductive data were scanty.

Turtles up to age 5 yr consistently showed complete sets of annually developed growth rings, and were aged by discriminating such rings from weak, non-annual rings (e.g., Tinkle, 1961). Some turtles up to age 9 yr also had complete sets of rings. In older individuals lacking some juvenile rings, the smallest annulus was used to calculate size at that age, and the growth trajectory shown by subsequent annuli was projected backwards to estimate time to hatchling size (Sexton, 1959; Zug, 1991). Eggs were assigned age 0, and hatchling age 1, because laboratory data show that eggs of this species undergo diapause and hatch approximately 11 mo after laying (personal observations, M. Ewert, personal communications).

Age at maturity in males was assessed by categorizing male secondary sexual characteristics (elongated tail, concave plastral shape, cornified patch on ankle) as immature, maturing, and fully mature for the smallest known males (those with sexual characteristics distinguishing them from females and unsexable juveniles). For females, age at maturity was determined by palping for eggs in known-age individuals.

Survivorship and population size were computed using the Jolly-Seber method (Krebs, 1989) with Program Jolly (Hines, 1992). Each year of sampling was defined as one sampling period, and the mean date of capture for all individuals in that year was used as the sampling date to compute sampling interval. This violates an assumption of the method, but not very severely, since inter-sample intervals were always longer than the interval encompassed within samples. Since the algorithms are too simple to cope with the extended age structure present in turtle populations, the data set was partitioned for analysis, and subsets combined when they showed no apparent variation in survivorship. For example, young adults (5-8 yr of age) were compared to only older adults, but these groups were lumped when no substantial difference in computed outcome was found. By contrast, 2 yr olds were analyzed by subsetting the dataset to include only their single cohort as it progressed through the yearly samples, this operation being performed separately for each cohort for which sufficient recaptures were available. Whereas 2 yr olds had lower survivorship than adults, 3-4 yr olds did not, or had only marginally lower survivorship. However, attempts to lump all animals aged 3 yr or older failed due to capturability bias detected by Program Jolly goodness-of-fit tests.

Among the models available with Program Jolly, Model A, with time-varying capturability and time-varying survivorship, satisfied goodness-of-fit criteria consistently, indicating low capturability bias. The appropriateness of Model A was as expected, considering that the dataset was assembled with annual variation in sampling effort, and considering the likelihood that survivorship and recruitment vary from year to year in this population. This model, however, computes mean survivorship by unweighted averaging of sampling period values, even though the sampling periods vary greatly in duration and quality of data. To avoid these anomalies, the means were recomputed on the computer printouts by weighting each computed value by the duration of the sampling interval. The results obtained in this way were consistent with Models B and D in computational cases where they met goodness-of-fit criteria.

As a further check, adult survivorship was also calculated by assuming a stable age distribution and constant age-specific adult survival rate. Hypothetical survivorship values were applied iteratively, starting from the known number of 5 yr olds. The value correctly predicting total adults (aged 5 yr or older) is equal to the average survivorship. This method was previously checked against long term recaptures at another study area (Rosen, 1987) and found to be acceptable. It is well-adapted for turtle studies, where number of young adults can often be readily determined by growth ring analysis, and the total number, but not the age, of all older adults can also be determined. In an earlier analysis (Rosen, 1987) it was noted that the age distribution at Quitobaquito was not stable, which may still be true. However, the longer study period available now for analysis helps smooth short-term fluctuations in age structure that would make the method questionable. This, and the lack of great change in population size over the duration of sampling, also support the idea that deviation from the stable age distribution was not serious enough to invalidate the method.

Additionally, population density was hand calculated for the many sampling periods during 1982-1985. Lincoln Index and Bailey triple catch methods were applied to the data, all results were weighted by sample size of recaptures involved in each computation, and a resultant weighted mean was computed.

RESULTS

Overall Sampling and Captures

We report on 560 records of Sonoran mud turtles at Quitobaquito, 1982-1995 inclusive. For mark-recapture computations, their were 211 marked individuals, with 484 captures (273 recaptures, of which 167 were suitable for a Jolly-Seber analysis using each year in which work was done as the sample unit). In addition to these, we marked 19 hatchlings ($\leq$ 3 mo post [August] hatching) and made five recaptures of these; and one additional recapture of the 19 marked hatchlings was made in the second year of life. In addition, we recovered carcasses or partial or complete shells of 22 individuals, of which 12 were clearly marked and 6 were clearly unmarked (6 shells were incomplete, and mark status could not be ascertained). In 1993, we obtained 24 location fixes on 5 radiotelemetered females; the relocations are not included in the summary numbers given above. The remaining records are additional hatchling observations which pertain, undoubtedly, to unmarked individuals.

Sampling effort and results are listed in Table 1. We trapped in 1982-5, 1989, 1992-3, and 1995, during a total of 74 days totalling 1099 trap-days for hoop nets, plus 470 trap days for hatchling (minnow) traps. In addition, we sampled from time to time using un-modified, unbaited minnow traps set on the bottom, although only fish were captured in these. Early in the study, and occasionally thereafter, we tried dipnetting from a boat at night, but the turtles proved too wary for this to be worthwhile, especially as such activity may interfere with trapping. In general, trapping success was usually low or moderate compared to expectations based on work at other localities.

Habitat Use Observations

Juvenile turtles were only in traps set at or near the vegetated margin of open water, or in dense emergent vegetation, whereas adults were found in all areas (see Fig. 2). Within the pond-margin habitat, juveniles were much more frequently captured along the edge of the dense stand of tules on the shallow shelf recommended by Lowe and Gloyd (Fig. 2). Juveniles constituted a large proportion of individuals trapped in the two main spring pools, early in the study (1983-1985) when extensive trapping was done there. Substantial numbers of juveniles, including many hatchlings, also were captured or observed in the channel that was constructed in 1989-90, as were a substantial number of adults. The preference of juveniles for shallower, more heavily vegetated water is as expected and widely seen (but rarely documented) in this and many other turtle species.

Adults were trapped in all areas of the pond, including open water, on the shelf (ca. 0.7-1.0 m depth) and in deep water (1.3-1.7 m depth), and around the pond margin. For deep water captures, traps near the shelf-deep water dropoff, and especially, at a large cottonwood projecting horizontally, half submerged, about 10 m into the pond, caught more turtles than those in structurally simpler habitat. Around the pond margin, trapping success for adults (and juveniles) was higher in areas with complex vegetation on and overhanging the bank, and lowest in areas with deep, steep banks having either tules or primarily woody vegetation.

Table 1. Turtle trapping for this project conducted at Quitobaquito, Arizona, 1982-1995. Number of traps set varied from 9-25 for hoop nets and 8-15 for minnow traps.

	Trap-Days:		
Dates	Baited Hoop Traps	Baited, Modifed Minnow Traps	Captures
1982:			
Sept. 19	13	0	3
Oct. 15 - 19	65	0	17
Nov. 16 - 17	26	0	11
Dec. 15 - 17	39	0	5
1983:			
Feb. 19	13	0	13
Mar. 15 - 17	39	0	5
Apr. 21 - 24	52	10	5
July 19 - 21	87	0	10
1984:			
May 15-17	42	8	16
June 5 - 7	67	0	7
July 28 - 29	38	22	7
1985:			
Mar. 15-16	40	26	4
Apr. 14	20	14	6
May 18 - 19	38	30	5
June 29 - 30	38	20	14
Aug. 9 - 11	48	45	12
Sept. 14 - 16	60	40	21
1989:			
Oct. 6 - 11	96	84	46
Nov. 2 - 5	64	56	19
1992:			
Oct. 27 - Nov. 1	96	88	82
1993:			
July 15 - Aug. 3	114	27	139
1995:			
Sept. 1	4	0	43
TOTAL	1099	470	490

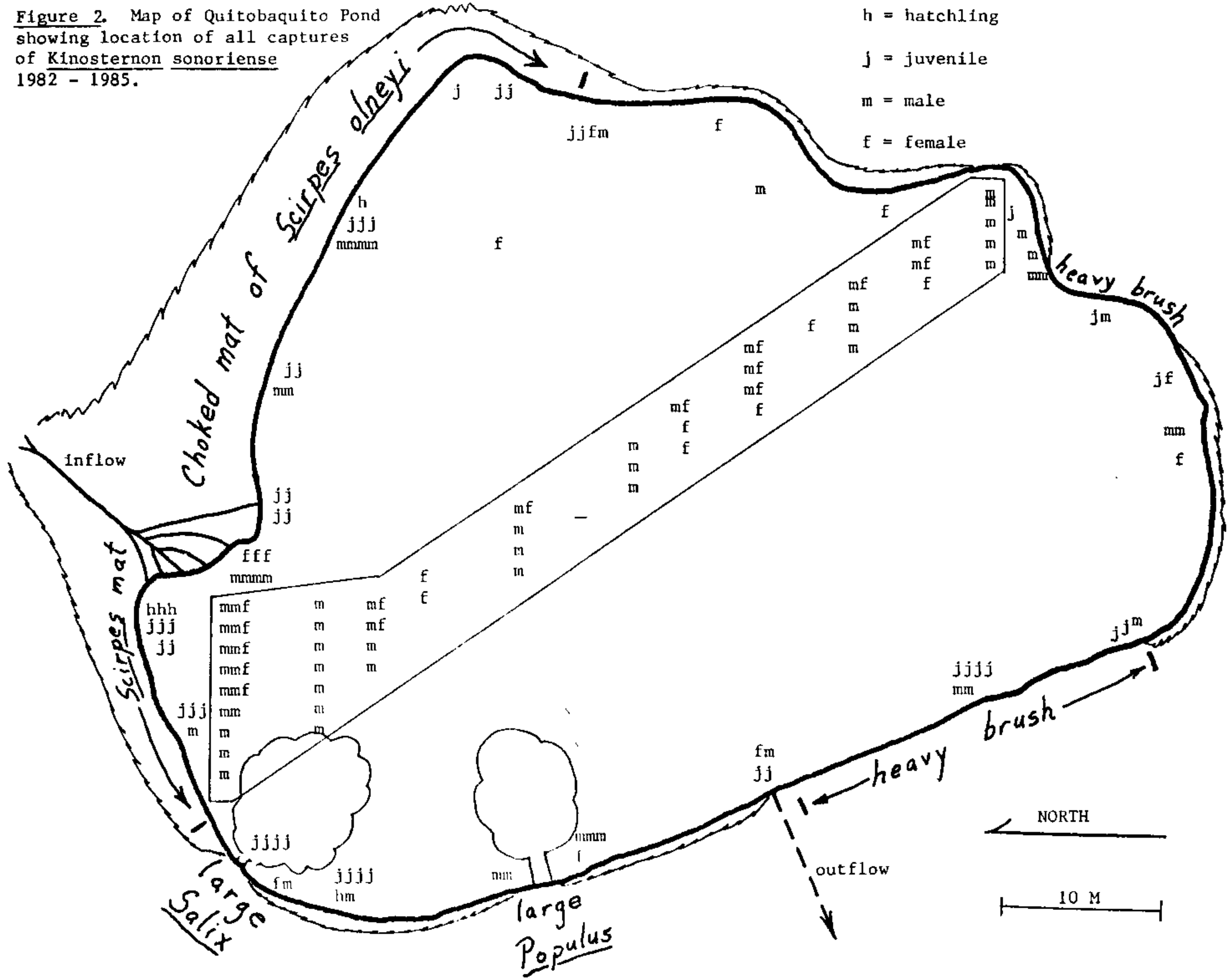


Figure 2. Map of Quitobaquito Pond showing location of all captures of *Kinosternon sonoriense* 1982 - 1985.

In 1992, baited hatchling traps that were set for 20 trap-days in the newly created spring channel yielded 14 juveniles. The capture rate significantly higher (3.3-fold higher) than in the pond (13 captures in 62 trap-days, $X^2 = 11.05$, $p < 0.05$).

In summary, juveniles strongly preferred shallow water with thick emergent vegetation; and trapping suggests that they used the semi-natural spring channel more intensively than the pond margins within at most 2 yr of its creation. Adults preferred areas with structural complexity in and above the water column, possibly regardless of depth. We noted no obvious seasonal trend in habitat use (i.e., no obvious avoidance of shallow water in summer).

Age, Growth, and Sexual Maturity

Size at age 7 at Quitobaquito was about 102 mm CL for females, the second lowest observed at any of 6 primary study sites for the species in Arizona (see Rosen, 1987), but rather similar to an apparently resource-limited population at Tule Creek, another small spring in Sonoran desertscrub (Rosen, 1987). The growth pattern is shown in Fig. 3. In females, size increase was most evident up to age 6 yr, reduced from age 7 yr to age 10-12 yr, and very slow thereafter. In males, size increase with age appears to slow markedly at age 4 yr, continue at a reduced rate to age 10, and continue very slow thereafter.

Immature males were 2-3 yr of age and 60-79 mm CL, while maturing males were age 3 yr and 75-80 mm CL; the smallest and youngest adult males were 84 mm CL and 4 yr of age (Table 2). This indicates a fairly consistent age at maturity of 4 yr in males, at 80-84 mm CL. The youngest female bearing eggs was at the end of her 5th year (approximate age 5 yr, 11 mo); dissection revealed that she had not previously ovulated. This female was also the smallest gravid individual, at 100 mm CL. This suggests that age at maturity in this population is 6 yr, or just slightly less, as in the other populations studied (Rosen, 1987). The first (and most marked) inflection point in the age-size graphs for both males and females (Fig. 3) corresponds closely to age at maturity.

Attempts to fit mathematical growth curves to turtle data (Wilbur, 1975; Dunham and Gibbons, 1990) have not fully succeeded because the inflection points are real: presumably two equations, applied astride the main inflection point, would resolve most of the difficulty; however, Rosen (1987) noted a transitional period in young adult female Sonoran mud turtles, during which there was a gradual shift of investment from growth to reproduction. If this interpretation is correct, growth equations, at least for females, would fail again at a second inflection point that is probably at 11 yr (personal observation).

The observed age structure of Sonoran mud turtles at Quitobaquito is shown in Table 3. Growth rings were not studied on turtles which were captured prior to July 1983 but not subsequently recaptured, and hence age determinations were not made. All of these turtles were of adult size. The category "older" represents individuals that were clearly old and un-ageable at first capture. Turtles were observed to reach at least age 23, and undoubtedly some were older than this. Juveniles were present in numbers in all samples starting in July 1983, when trapping

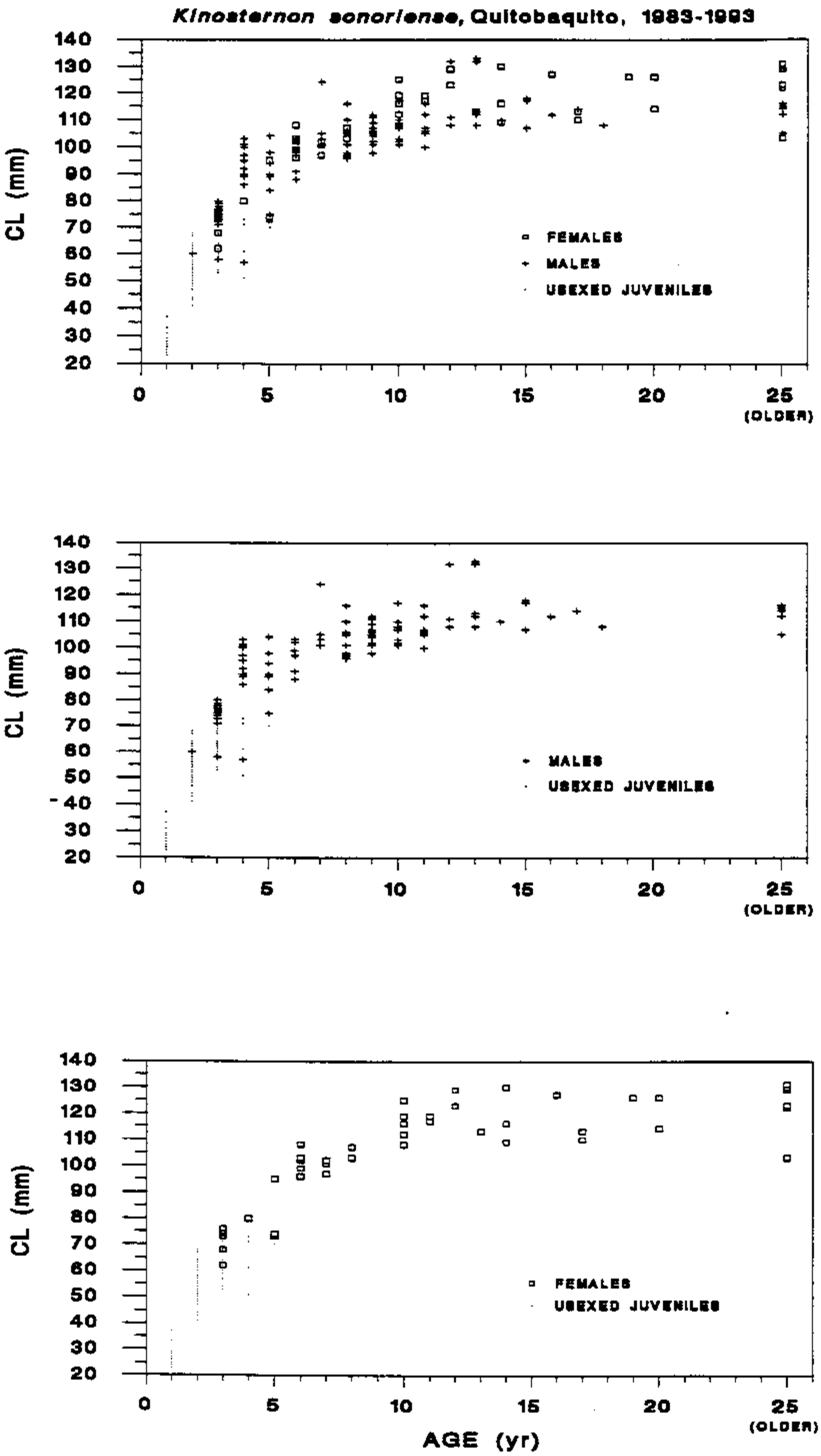


Figure 3. Age-size model of growth.

Table 2. Maturation data for male *Kinosternon sonoriense* at Quitobaquito, Pima County, Arizona. Categories are based on field determination by principal investigator.

		CL (mm):			Age (yr):		
	N	mean	min	max	mean	min	max
IMMATURE	4	70.7	60	79	2.8	2	3
MATURING	5	77.4	75	80	3	3	3
YOUND ADULT	5	87.2	84	89	4.83	4	6

Table 3. Age structure of the Sonoran mud turtle at Quitobaquito Springs, Arizona, 1982-1995. Individuals were counted once for each year in which they were observed. Those captured during 1982 - March 1983 were adult-sized, but were not aged unless subsequently captured. Other individuals that were too old to age at first capture are in the category "older". Sex was undetermined for young juveniles.

	AGE (YEARS), HATCHING = AGE 1																						
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	23	undet. older	undet. adult	TOTAL
FEMALES	0	1	9	5	10	4	5	3	4	6	5	3	1	3	0	1	2	0	1	1	1	10	14	89
MALES	0	0	14	13	8	15	9	14	16	8	7	3	5	1	3	1	1	1	1	0	0	9	18	147
Undet.	25	72	23	11	8	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	142
TOTAL	25	73	46	29	26	20	14	17	20	14	12	6	6	4	3	2	3	1	2	1	1	19	34	378

Table 4. Age and sex of Sonoran mud turtles found dead at Quitobaquito, Pima Co., Arizona, 1983-1995. The 4 adult females that died were carrying transmitters.

	AGE (yr)										adult, age	
	1	2	3	5	6	9	12	13	15	20	unk.	TOTAL
FEMALE			1		1		1			1	1	5
MALE				1	3	1		1	1		2	9
sex undet.	1	1	1	1								4
ALL	1	1	2	2	4	1	1	1	1	1	3	18

around the pond margin was initiated. Assuming a stable age distribution, the data in Table 3 yield a value of $s_x = 1 - q_x = 0.873$ (where x = age) for average annual adult survivorship.

Survivorship and Population Size

Jolly-Seber computations yielded mean values for mean annual adult survivorship of $s_x = 0.864 \pm 0.0435$ (SE) for Model A, 0.873 ± 0.0192 (SE) for Model B, and 0.890 ± 0.0186 (SE) for Model D, all in close agreement with the value in the preceding section derived from the age distribution. Goodness-of-fit tests suggest that Model A (temporally varying survivorship and catchability) best fit the data, with Model B (only catchability varies with time) a close second, and Model D (constant survivorship and catchability) a poor fit. This is intuitively satisfying, in that sampling was not uniform across the years of study, and survivorship probably varied, but varied relatively slightly in relation to the mean, over time. For adult plus subadult males and females (5 or more yr of age) under separate computation, only Model A gave acceptable fit: for males $s_x = 0.901 \pm 0.1719$ (SE), and for females $s_x = 0.855 \pm 0.0469$ (SE). Obviously, the computer program indicates that the sexes have indistinguishable survivorship rates, but the computed values appear to agree with the age structure, and suggest that female survivorship is slightly reduced. The consistency of all of the survivorship computations indicates that this population has modest or low adult survivorship compared to most others studied (Rosen, 1987).

Juvenile survivorship was computed by the Jolly-Seber program by stratifying the data set in time so that only one cohort of 2 yr olds (from sampling period 1 in the computation) was utilized at a time. Thus, one way to evaluate the performance of the program was to ensure that computed recruitment was close to zero, since no recruitment could occur into the closed cohorts so defined. The following four computations were performed: 1984 2 yr olds, $s_x = 0.54$; 1985 2 yr olds, $s_x = 0.70$; 1989 2 yr olds, $s_x = 0.85$; 1992 2 yr olds, $s_x = 0.64$. Only Model A met the goodness-of-fit criterion. The mean is $s_2 = 0.68$ and is probably inflated because 1985 2 yr olds were not resampled until 1989, and 1989 2 yr olds were not resampled until 1992--thus higher survival at greater ages (next paragraph) would have been lumped into the computation. I therefore arbitrarily set $s_2 = 0.62$. There is convincing evidence that survival from age 2 to 3 yr is lower than adult survival.

For older juveniles (initial ages 3 and 4 yr), computations could only be performed by combining the 2 cohorts into single, no-recruitment strata within the datasets. Three resultant computations yielded a mean of $s_x = 0.84$ for older juvenile survival. At these ages, it is quite possible that adult survival rates were attained. However, slightly elevated mortality is reasonable *a priori* based on smaller size of the turtles, and the computed value was accepted for use in demographic calculations.

Age distribution of mud turtles found dead at Quitobaquito (Table 4) suggests the possibility that older juveniles and young adults may have higher mortality than older adults. The small number of young juveniles in Table 4 probably results from consumption, shell and all, of young turtles by predators and scavengers. Table 4 suggests a survivorship curve that does not reach the adult maximum until age 6 or 7 yr. The ratio of adjacent age classes in Table 3 suggests $s_2 = 0.63$, $s_3 = 0.63$, $s_{4+5} = 0.83$. These values support or confirm the computations

Table 5. Population estimates for the Sonoran mud turtle at Quitobaquito Springs, Organ Pipe Cactus National Monument, Arizona, based on data from 1982 - 1995. Hatchlings are not included in any of the estimates. Estimate is accompanied by 95% confidence interval, in parentheses, where available.

	Estimated Population Sizes				
YEAR	Adults (5+yr)	Age 3-4yr	Age 2yr	Total	Source and method
1971	-	-	-	143 (91-195)	Gehlbach, Lincoln Index.
1983-4	67 (41-93)	12 (-)	29 (-)	108 (71-158)	This study, Lincoln Index, Triple Catch.
1983-4	66 (-)	16 (-)	30 (-)	-	This study, Program Jolly.
1985	61 (28-95)	13 (-)	40 (14-60)	114 (-)	This study, Program Jolly
1989	49 (29-70)	17 (-)	14 (5-23)	68 (36-101)	This study, Program Jolly.
1992	66 (43-90)	22 (-)	42 (31-53)	115 (71-160)	This study, Program Jolly.
1993	70 (44-96)	39 (24-49)	24 (18-30)	134 (86-183)	This study, Program Jolly.

described above, with the lower value for s_3 possibly a result of a change in capturability between ages 3 and 4, as well as a lower survivorship at age 3 yr that might be expected based on consideration of size and shell hardness. Again, a gradual transition from lower to higher survivorship is indicated, rather than an abrupt and early transition to the relative safety of adult existence. In these latter computations, the intensive sampling, and the procedure of counting of an individual in Table 3 only once for each year in which it was observed, probably overrides the otherwise-expected age-related capturability bias.

Program Jolly yielded population size estimates for the three age classes defined in the computational procedure (Table 5). Program Jolly's estimates are similar to those derived from Lincoln Index and Triple Catch methods for the 1983-1985 data (Table 5). The adult population appears to have remained fairly stable over the period 1983-1993, although there was probably a small decline during the late 1980's. The 2 yr old population estimate was low for 1989, and that for older juveniles (aged 3-4 yr) was highest in 1992 and 1993. The data set is probably too small to trust the differences among years in Table 5, although the numbers of juveniles in 1989-1993 appear to be consistent with the results expected based on numerous within-year recaptures during the intensive and successful sampling in those years. Lincoln Index values computed by hand for 1989, based on within-year recaptures, also indicated a lower number of adults than similar computations for earlier years. Overall, the data suggest a decline from 1971 to 1989, followed by a possible increase in the early 1990's.

Reproduction

Four of 12 females palpated for eggs during the egg-carrying season (July 19-September 12) were gravid. This proportion gravid corresponds to average production of 1.4 - 1.6 clutches per females per year, based on more in depth study of other populations (Rosen, 1987). A mean value of 1.5 clutches per year was assumed, and it was assumed that only a single clutch would be produced in the first adult year, as suggested by more abundant data for other populations (Rosen, 1987).

Data are available for 3 clutches based on X-ray, dissection, and oxytocin-induced laboratory oviposition (Ewert and Legler, 1978). Clutch size averaged 4.0 eggs, and the largest female had the largest clutch and largest eggs, the pattern expected in this species (Hulse, 1974a, 1982; Rosen, 1987; personal observations). Relative clutch mass averaged 0.069, suggesting that clutch mass in this population is low, as in about half the studied populations (Rosen, 1987), and egg mass at 4.9 gr/egg is moderately high for turtles of the sizes shown in Table 6.

Among five adult females followed intermittently with radio transmitters during August - mid-September 1993, 3 were definitely known to have produced eggs. Beginning in the third week of August, an attempt was made to capture each female weekly for palpation to check for eggs. Over the 4 weeks of this operation, there was difficulty capturing turtles (averaging half an hour of muddling [probing the pond bottom by hand] for capture), and 4 of the 5 females died, all apparently within 0-2 days after an episode of capture and handling. It is not possible to infer clutch frequency from these data.

Table 6. Female reproductive data for *Kinosternon sonoriense* at Quitobaquito, Pima County, Arizona, 1983-1995. RCM is clutch mass/female mass (including eggs).

		Female:			Eggs:			Clutch:		
DATE	DATA TYPE	AGE (yr)	CL (mm)	MASS (gr)	LENGTH (mm)	WIDTH (mm)	MASS (gr)	# EGGS	MASS (gr)	RCM
15-Aug-93	dissection	6	102	195	29.7	15.6	4.22	3	12.67	0.065
22-Jul-83	X-ray	12	119	274	30.2	17.2	5.03	3	15.09	0.055
29-Jul-84	oxytocin	>12	129	378	30.6	17.5	5.40	6	32.40	0.086
MEAN			116.7	282.3	30.17	16.77	4.883	4.0	20.053	0.0687

The physiological cause of death for these females is unclear. Stress of staying underwater for at least half an hour, along with the fear experienced, and the poor nutritional state of the animals (below) may have been involved. The turtles were apparently alive when released, and in no case was the transmitter (1.0 g, encased within a smoothed glue layer) caught on vegetation. Dissection of one of the 3 salvageable carcasses revealed atretic follicles in early September, and indicated possible reproductive failure for the year. The other two dissections revealed fresh corpora lutea or oviducal eggs, and in both cases 3 or more ovulatory-sized ovarian follicles. Fat pads were small in all 3 females.

One of the radio-tracked females was found apparently preparing to lay eggs by burying herself about 15 cm into the soil in mesquite bosque 9 m WSW of Quitobaquito Pond, on September 8, 1993. Unfortunately, this female was dug up, and then not tracked; she subsequently deposited eggs within four days or less at an undetermined place. Other females were not observed during nesting; non-gravid females were not found on land, although one radio-tracked female was found buried in mud among tule roots along the north side of Quitobaquito Pond.

Sex Ratio

For turtles $\geq$ 5 yr of age, at total of 69 individual males and 44 females were recorded. Program Jolly computations consistently produced mean estimates of 40-42 males and 19-21 females. The adult sex ratio is even more skewed than indicated, since males mature a full year earlier than females. A value of 2:1 for the adult sex ratio is indicated. The calculated difference in mean adult survivorship of 90% for males to 85% in females implies a 1.59:1 sex ratio if the sexes were equally numerous at age 5. Even, if this is so, there still remains an unaccounted for 1.26:1 imbalance in the sex ratio (it is unclear exactly which data values should be used to test goodness-of-fit, but regardless, $X^2 = 1.03 - 2.40$, $P > 0.1$; this unaccounted for imbalance may be insignificant).

An examination of the distribution of hatching years for Quitobaquito turtles (Table 7) suggests that the sexes may be produced differentially in different years. Table 7 should be interpreted cautiously, because data for 1983-1993 are inherently male-biased throughout. This is so because males can be sexed at a younger age than females, and this bias is magnified by the large number of 2 and 3 yr olds in the samples. Nonetheless, the period approximating 1974-1978 appears to have been one yielding high male recruitment, as well as a good time for recruitment overall. There may have been an increase during the 1990's in the proportion of females recruiting, but sample sizes remain too small to confirm such a trend.

At this warm, xeric margin of the geographic distribution, and given the very high temperatures of the mid 1980's through 1995, sex ratio anomalies in turtles may be expectable, since egg incubation temperature usually determines sex (Ewert and Nelson, 1991). Ewert and Nelson (1991) found that in species with females averaging larger than or similar to males in size, as in the Sonoran mud turtle (Hulse, 1974a; Iverson, 1988), low incubation temperatures (ca. 27°C at the critical time) yield males and higher temperatures yield females; subsequent work confirmed this for the Sonoran mud turtle (M. Ewert, personal communication). The period producing high male recruitment at Quitobaquito was relatively cool, with low rainfall and moderate drought conditions (Sellers et al., 1985), tending to support a hypothesis that the observed sex ratio might be a result of, at least partly, climatic fluctuation or change via the effects of incubation temperature. The kinds of data needed to resolve this possibility are currently unavailable, but it remains an important hypothesis for conservation, and is of interest to chelonian biology.

Recruitment Patterns and Early Survivorship

Assuming that the estimated average of 20 adult females each produced 1.5 clutches/yr of 4.0 eggs/clutch, there would be about 120 eggs per year produced during the study period of 1983-1995. There was no evidence of nest predation, of the kind often seen in turtles, and we therefore assumed that nest predation was negligible. Based on assumptions of low nest predation and high egg fertility, we assigned a value of 90% for hatching success (under laboratory conditions, at least 81% of Sonoran mud turtle eggs obtained by oxytocin injection may hatch [M. Ewert, personal communication], and embryonic viability may be 85-100% for turtles under good conditions [Ewert, 1985]). This gives an estimate of 108 hatchlings per year, which we used for further demographic analysis. The assumption of 90% hatching rate affects the estimate for first year survivorship, but does not affect subsequent survivorship estimates or the life table computations.

Sampling of hatchlings produced a very limited number of recaptures. Five hatchlings were marked in 1985, and 9 in 1989, but there was no sampling the following years, and marks on these turtles may have disappeared; none were recaptured. Five were marked in 1992, and one of these was recaptured among the seventeen 2 yr olds observed in 1993. This (5 x 17 = 85 hatchlings) is roughly congruent with assumptions about reproductive output and hatching success. Within seasons, 3 recaptures yielded three Lincoln Index estimates averaging 15 hatchlings (9 - 21), but this low value is expected on the short intervals between recaptures, the low mobility of hatchlings, and the limited extent of trapping in spring and channel habitats. Assuming 90% hatching success, there would be 108 hatchlings per year; the mean value for 2 yr old cohorts of 30.3 individuals (Table 5) yields a survivorship estimate through the first year after hatching at $s_1 = 30/108 = 0.28$. A low value in this range is not unexpected based on the array of predators capable of consuming the small, soft-shelled, hatchlings.

Table 8 suggests temporal fluctuation in successful recruitment of Sonoran mud turtles into the adult population at Quitobaquito. In the table, 1-2 yr olds were not counted, in order to reduce the bias caused by sampling less than every year in a population with a large crop of 2 yr olds. Identical trends are revealed by all the data, but the table is much more difficult to interpret. The pulse of good recruitment in the early-mid 1970's, described above, is also evident in Table 8. There was also apparently good recruitment during the 1982-1985 period of average or wet years, and again in 1989, following good and timely rains in 1988 when eggs for 1989 hatchlings were produced. The depths of drought in 1989-early 1990 resulted in depressed recruitment in 1990, but the strong rains of summer 1990 and moderate rains of the following winter apparently led to a bumper crop of 1991-hatched recruits. For the recent period, recruitment was correlated with rainfall during the recent past. The foregoing period of high

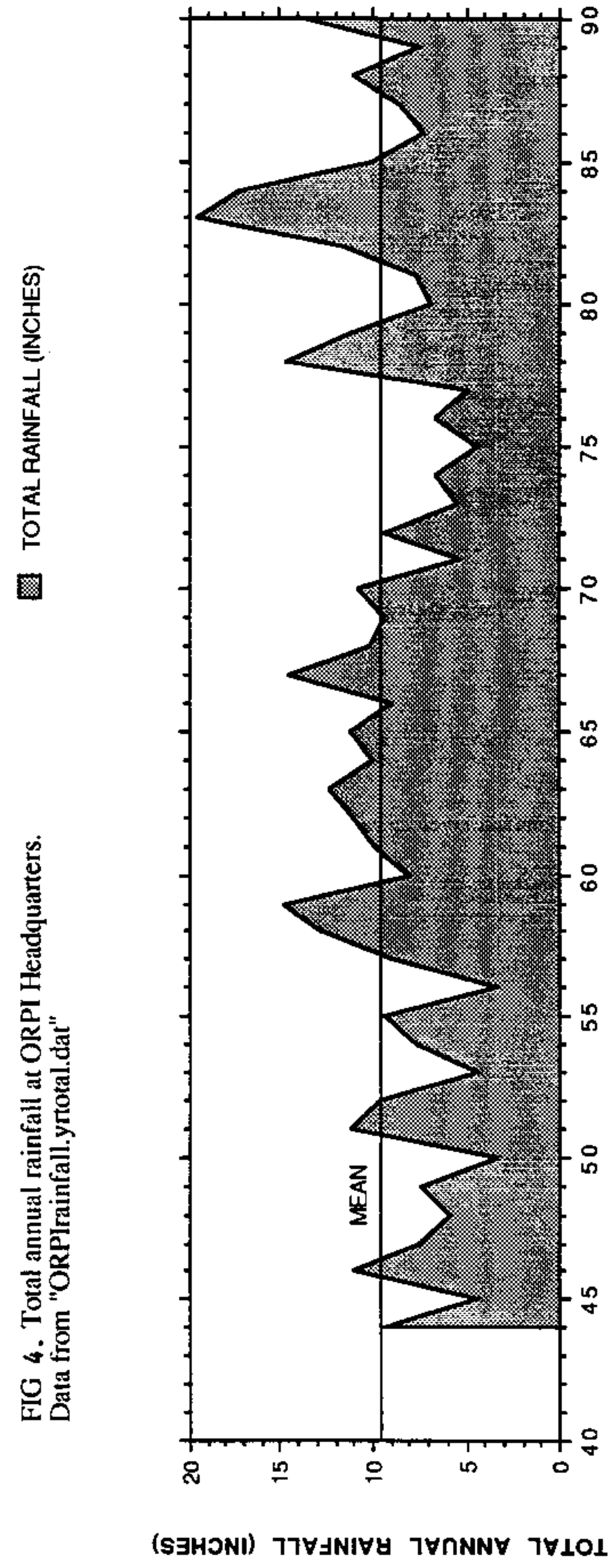


FIG 4. Total annual rainfall at ORPI Headquarters. Data from "ORPIrainfall.yrtotal.dat"

Table 7. Estimated year of hatching based on growth rings for the Sonoran mud turtle at Quitobaquito, Arizona, based on sampling during 1982-1995. No estimate was available for individuals too old to read growth rings when first captured. Eggs are thought to hatch one year following oviposition. Sex usually could not be determined in young juveniles not recaptured at or near maturity. Each individual observed during the study is counted once.

	YEAR OF HATCHING:																						
	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86	87	88	89	90	91	92	93	undet.	Total
FEMALES	3	2	0	2	1	1	0	0	1	0	0	2	1	2	2	1	1	1	1	4	3	1	17	46
MALES	0	2	3	7	5	8	2	1	1	2	3	3	2	4	5	3	1	5	2	4	2	0	18	83
undet.	0	0	0	0	0	0	0	0	0	1	4	10	14	6	1	1	8	14	8	30	21	1	1	120
TOTAL	3	4	3	9	6	9	2	1	2	3	7	15	17	12	8	5	10	20	11	38	26	2	36	249

Table 8. Estimated year of hatching as in Table 7. Young juveniles (<3 yr of age) are excluded. Each individual is counted once for each year in which it was captured.

	YEAR OF HATCHING:																						
	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86	87	88	89	90	91	92	93	undet.	Total
Sample Year:																								
1982	1	2	1	2	3	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	17	30
1983	1	3	1	8	3	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	10	29
1984	0	1	3	3	1	4	1	1	0	2	4	0	0	0	0	0	0	0	0	0	0	0	11	31
1985	1	2	1	3	2	3	1	1	1	0	3	5	0	0	0	0	0	0	0	0	0	0	2	25
1988	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	1
1989	0	1	1	2	3	2	1	0	1	0	1	0	2	2	4	2	0	0	0	0	0	0	4	26
1990	0	0	0	0	1	0	0	0	1	0	0	0	0	1	1	0	0	0	0	0	0	0	1	5
1992	0	1	1	1	1	1	0	0	0	1	0	2	3	2	5	4	1	5	7	0	0	0	2	37
1993	0	0	0	0	0	1	0	0	0	2	0	1	2	2	2	1	4	8	2	26	0	0	2	53
1995	0	1	0	0	0	1	0	0	0	0	0	0	1	0	1	2	2	2	2	13	12	2	4	43
All Years	3	11	8	19	14	17	4	3	3	5	8	8	8	8	13	9	7	15	11	39	12	2	53	280

Table 9. Age structure of the Sonoran mud turtle at Quitobaquito Springs, Arizona, during sampling "eras" in the 1982-1995 study. Rationale for the eras is explained in the text. Individuals were counted once for each year in which they were observed. Those captured during 1982 - March 1983 were adult-sized, but were not aged unless subsequently captured. Other individuals that were too old to age at first capture are in the category "older". Sex was undetermined for young juveniles.

Tine of Sample	AGE (YEARS), HATCHING = AGE 1 1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	23	undet. older	undet. adult	TOTAL
82 - early 83	0	0	0	0	0	5	4	4	5	2	3	0	0	0	0	0	0	0	0	0	0	6	21	50
mid 83 - 85	5	22	9	5	1	2	2	6	8	6	7	3	2	1	0	0	0	0	0	0	0	4	9	92
88 - 90	15	8	2	5	3	3	0	1	0	1	1	1	2	3	3	1	1	0	0	0	0	4	1	55
92 - 95	5	43	35	19	22	10	8	6	7	5	1	2	2	0	0	1	2	1	2	1	1	5	3	181
Full sample	25	73	46	29	26	20	14	17	20	14	12	6	6	4	3	2	3	1	2	1	1	19	34	378

Table 10. Yearly breakdown of Sonoran mud turtles found dead at Quitobaquito, Pima Co., Arizona, 1983-1995.

	YEAR FOUND					
AGE (yr)	84	89	90	92	93	TOTAL
1		1				1
2		1				1
3		1			1	2
5		1	1			2
6	1		2		1	4
9					1	1
12					1	1
13					1	1
15			1			1
20					1	1
adults, age undet.		1		1	1	3
age undet.			4			4
TOTAL	1	5	8	1	7	22

recruitment, however, occurred during a relatively dry period of the 1970's (Fig. 4), although not long after refilling the pond in 1970 (Bennett and Kunzmann, 1989).

Age structure is subdivided among sampling "eras" of the 14 year study in Table 9. The relatively large samples of 1983-1985 and 1992-1995 differ markedly from one another in the proportion of older juveniles. In the earlier period, 2 yr olds were 23.9% of the sample, and remained at 23.8% 10 yr later. In contrast, 3-5 yr olds composed 16.3% of the observed sample in 1983-1985, significantly below the 42.0% observed 10 yr later ($X^2 = 14.58$, $p < 0.05$). There was significantly higher recruitment into the subadult age class in the 1990's. For the smaller sample from 1989 trapping, 17.8% of the turtles were in the subadult class (ages 3-5 yr), and an equal number (n = 8) were aged 2 yr, suggesting demographics similar to that seen in the previous sampling interval. The age structure data suggest increased juvenile survivorship in the 1990's.

Observations of Mortality, and Hypotheses of Causation

It is rare to find dead Sonoran mud turtles in the wild, and those found usually show signs implicating predation as the likely cause of death (Rosen, 1987; personal observations). The 22 dead turtles observed at Quitobaquito from 1982-1995 (Tables 6, 11) represent an unusual number considering the small population size. None showed unmistakable evidence of predation. For most cases when fresh carcasses were found, there was no evidence at all to indicate cause of death. Inexplicably dead turtles were found to be marked or unmarked, recently trapped or not, and (Table 5) were of various demographic groups. The peak of observed, unexplained mortality was in late 1989 and early 1990 (Table 10), which was the period of most severe drought during our study period, and also included record heat throughout southern Arizona in mid-June 1990.

The appearance of turtle carcasses is a signal that something may be amiss (e.g., Bishop et al., 1991). We investigated the possibility that the unexplained mortality was caused by (1) pesticide intoxication, (2) heavy metal poisoning, (3) hypersalinity, or (4) dietary deficit of energy. Eight carcasses of mud turtles that died at Quitobaquito were available for analysis; these were collected 1990-1993 in relatively fresh condition, subjected to gross dissection, frozen, and sent via USFWS to Hazelton Environmental Services, Inc. (525 Science Dr., Madison, WI, 53711) for chemical analysis (Environmental Protection Agency, 1986). Water samples were taken in 1989 (by P. Rosen); and sediment samples were collected in 1994 (Kirke King and Cynthia Martinez) and sent with the carcasses for chemical analysis.

Results of the carcass and sediment chemical composition analysis are described and discussed in detail by King et al. (1996), included here as Appendix B. In the present section, we focus on highlights of that work.

Hazelton Environmental screened sediment samples and homogenates of each carcass for the following organochlorine compounds: PCB; alpha-, beta-, and gamma-BHC; alpha- and gamma-chlordane; oxychlordane; dieldrin; endrin; heptochlor epoxide; mirex; toxaphene; trans-nonachlor; and o,p'- and p,p'-DDD, DDE, and DDT. The only positive findings were for 4 of the turtles with p,p'-DDE, a metabolite of DDT, at levels just above the detection limit, averaging

Table 11. Chemical composition at Quitobaquito. Concentrations are given in ppm (mg/l). Included are total dissolved solids (TDS) and total organic carbon (TOC). Values are means based on various sample sizes. Sediment and carcass values are ppm-dry weight.

SOURCE	Cole and Whiteside (1965)		Bennett and Kunzmann (unpubl.)	Fisher (1989)	Rosen, K. Young (this study)		
YEAR	1963	1964	1981 - 1983	1984 - 1985	1989	1994	1989 - 1993
MEDIUM	water	water	n=1 to n=37 water	water	n=3, water	n=4, pond sediment	n=8 turtle carcasses
TDS	ca. 800	ca. 1300	688	ca. 700	952		
TOC			3.3				
pH		7.7		7.9 - 9.7			
Cations:							
Na	191.0	317.0	214.9				
K	4.5	6.5	4.5				
Ca	34.0	32.0	20.9				
Mg	12.6	14.6	9.9			14783.0	2055.7
Fe	0.09	0.08	0			6273.1	93.2
Sr						1166.2	868.8
Anions:							
Cl	148.0	350.5	160.5	134.6			
Si	28.0	42.0	21.4				
F		4.3	5.5				
HCO3	316.0	406.0	213.8	425.8			
SO4			98.9	99.3			
NO3	2.60	1.10	9.50	1.37			
PO4	0.000	0.000	0.000	0.012			
Metals:							
Al						5942.1	66.8
As			0.01			15.23	0.34
B						35.9	71.6
Ba						102.9	81.9
Be						0.5	0
Cd						0	0
Cr						55	3.95
Cu	0.1	trace	0.04			8.1	4.6
Hg			0			0.071	0.092
Mn						88.7	3.35
Mo						8.7	0.0
Ni						4.9	3.1
Pb			0.2			9.2	0.0
Se			0.04			4.9	1.9
V						130.9	0.2
Zn			0.05			20.9	169.1

0.0455 ppm dry-weight basis. These levels are probably not high enough or consistent enough to support a pesticide contamination hypothesis for the mortality (e.g., Hoffman et al., 1995). In addition, Mike Kunzmann (personal communication) tested a single carcass found at Quitobaquito in 1982, finding a low, presumably non-toxic concentration of the herbicide Dacthal (dimethyl tetrachloroterephthalate). These results tend to rule out pesticide intoxication as a cause of mortality, but they do not do so entirely. These pesticides bioaccumulate in lipid (body fat) and may be released into metabolic pathways during episodes of food scarcity when an animal is utilizing stored energy. Lipid storage was scant in mud turtles at Quitobaquito, rendering pesticide detection difficult, but not necessarily reducing the release of toxins during lipid utilization. It should be pointed out that detection of pesticides in four of the turtles, under the circumstances, is remarkable enough: it is not clear why any such amounts should be present in the system to begin with.

Table 11 shows the inorganic composition of Quitobaquito water, sediment, and turtle carcasses. The most suspect metal in Quitobaquito water or sediment is arsenic: one might prefer not to drink large quantities of the water for very long periods of time (see Peters, 1955; Tseng, 1977), but all the levels observed are far below known toxic concentrations in other animals (McGeachy and Dixon, 1990; Woods and Fowler, 1977, 1978; Goering et al., 1987; Jelinek and Corneliussen, 1977). None of the other metals or ions are at unusual concentrations (Table 11; see e.g., Hoffman et al., 1995; Prosser, 1973). We were unable to locate published data specifically on turtles that would assist interpretation of the elements involved; nonetheless, the possibility of direct metal poisoning appears to be remote. King et al. (1996; see Appendix B herein) presented comparative data for softshell turtles indicating that some elements (boron, chromium, selenium, strontium, and zinc) may be at especially high levels in the turtle carcasses from Quitobaquito. They could not establish toxicity on this basis, however. Arsenic and nickel levels were also higher at Quitobaquito than in the Gila River softshells, and copper and mercury were at lower levels. King et al. (1996) did not consider element concentrations in carcasses from Quitobaquito normal, and suggested the possibility of interaction between dietary insufficiency (below) and chemical toxicity.

The pond is relatively saline, with total dissolved solids (TDS) varying from 688 ppm to at least 1300 ppm, as in many western waters (e.g., Cole, 1983). Even if higher concentrations occurred, these would not remotely approach blood (or seawater) concentrations (Eckert et al., 1988; Cole, 1983), and would not be expected to cause physiological disorder or stress in the turtles. As noted by Fisher (1989), the inorganic and autotrophic components of the Quitobaquito ecosystem appear to be in good condition.

Gross dissection showed the turtles to be low in visible stored lipid (fat pads). Generally, fat pads were small, or even completely collapsed (i.e., probably containing no stored lipids), and livers were small, averaging only 0.32% of wet body mass. The livers looked unhealthy, although decomposition had been initiated by the time the turtle carcasses were collected and chilled. One of the three adult ovaries examined (above) also appeared to be lipid-poor, while the other two had several yolked follicles. Mortality of these individuals was proximally connected to stress of capture and handling. Lipid extractions (Soxhlet extractor with hexane solvent for non-polar [storage] lipids) from the 8 carcasses confirm a low level of body fat ($\bar{X}$ = 2.81% lipid wet-weight basis, s.d.= 1.038, range 0.89-4.18. The lipid index (100 X g fat/g lean dry mass) at Quitobaquito was also low (X = 7.6% ± 3.71 s.d.). These turtles appeared leaner than others dissected from a less resource-limited population (Rosen, personal observations). The female with atretic follicles and no visible lipid in the fat pads (above) had by far the lowest lipid content (0.89%).

Comparative data on body lipids in turtles is not abundant in the literature, but what is available confirms the low nutritional state of turtles at Quitobaquito. A variety of methods have been used in lipid analyses of turtles, leading to errors that may be as large as 17.5% overestimation (Dobush et al., 1985); this magnitude of error still permits some general conclusions to be drawn. Low fat values (LI = 4 - 10%) reported for box turtles and Texas tortoises (Brisbin, 1972; Rose, 1980) are exceptional rather than the norm. Congdon and Tinkle (1982) reported a fat female midland painted turtle at 26% lipid on a dry-weight basis (LI = 35%). Pond (1978) reported a value of 6% lipid wet-weight basis for a single snapping turtle, and Brenner (1970) reported percent fat in various tissues of wood turtles at 8.3-36.7% wet-weight basis. Data for 14 spiny softshell turtles (*Apalone spinifera*) from the lower Gila River, Arizona, give X = 16.47 ± 4.128 s.d., range 11.3-23.8% lipid wet-weight basis (Kirke King, John Moore, unpublished data).

Mud and musk turtles may often have substantial lipid reserves. Rose (1980) and Long (1985) reported very high lipid index values (ca. 20-40%) for the yellow mud turtle, and results presented by McPherson and Marion (1982) for the common musk turtle also indicate lipid index values in the range of 15-20%. Belkin (1965) presented data for 4 freshly captured loggerhead musk turtles X = 6.4% fat wet-weight basis, and 4 individuals that starved to death had X = 2.42% ± 0.974 s.d. (range 1.4-3.6%) fat wet-weight basis. Belkin's (1965) values for starved turtles is very close to the 2.81% fat observed in the dead turtles at Quitobaquito. Why turtles should starve with this much fat remaining is unclear (Belkin, 1965), but several scenarios relating to nutritional stress can easily be envisioned. The available data suggest that Sonoran mud turtles at Quitobaquito were markedly undernourished, contributing importantly to their mortality.

Food Availability and Diet

Sonoran mud turtles are omnivorous, feeding on fish and other vertebrates, aquatic invertebrates of all kinds, and plants (Hulse, 1974b). They consume fish avidly, but are not efficient at capturing them (personal observations), and vertebrates comprise a small proportion of the realized diet (Hulse, 1974b). The greatest part of the diet is invertebrates, especially benthic or vegetation-living forms, but herbivory is important in resource-poor environments (Hulse, 1974b). At Quitobaquito, dissections and feces from live turtles showed large amounts of plant material, primarily young shoots of tule (P. Rosen, personal observations).

Modified minnow trap data yielded comparative data on invertebrate abundance at Quitobaquito in different seasons and years (Fig. 5). Invertebrates recorded in these traps most frequently were larger predaceous kinds such as predaceous diving beetles (*Dytiscus* spp.), bellastomatids (*Bellastoma, Lethocerus*), odonate nymphs (Anisoptera), water scorpions (*Ranatra*), naucorid bugs, as well as water scavenger beetles (Hydrophilidae). Additionally, fishing spiders (c.f., *Dolomedes*) and water striders (Gyrinidae) occurred uncommonly. Sonoran mud turtles feed on these taxa (Hulse, 1974b), and these large and often predaceous arthropods probably reflect the underlying abundance of smaller invertebrates also consumed by turtles.

Methodology identical to that used at Quitobaquito (including the same traps and trapping protocol) was used to sample aquatic invertebrates at other Sonoran mud turtle population sites in Arizona. Arthropod abundance was low at Quitobaquito, although not as low as at Sharp Spring, Santa Cruz County (Fig. 5). In the laboratory, adults from the Sharp Spring population (but not other populations) occasionally died, probably from nutritional stress (personal observations). Over all populations with comparable invertebrate trapping data (Fig. 6), growth rate correlates closely with invertebrate abundance. More haphazard sampling of invertebrates at other sites also strongly supports the trend in Fig. 6. On some trips to Quitobaquito, invertebrate abundance fell to 0.19 (October 1989) and 0.17 (October-November 1992) invertebrates/trap day, well below the value recorded at Sharp Spring. Dead turtles were found floating in the pond in fall of 1989 and 1992, again suggesting that stresses associated with poor nutrition are important contributors to observations of unexplained mortality.

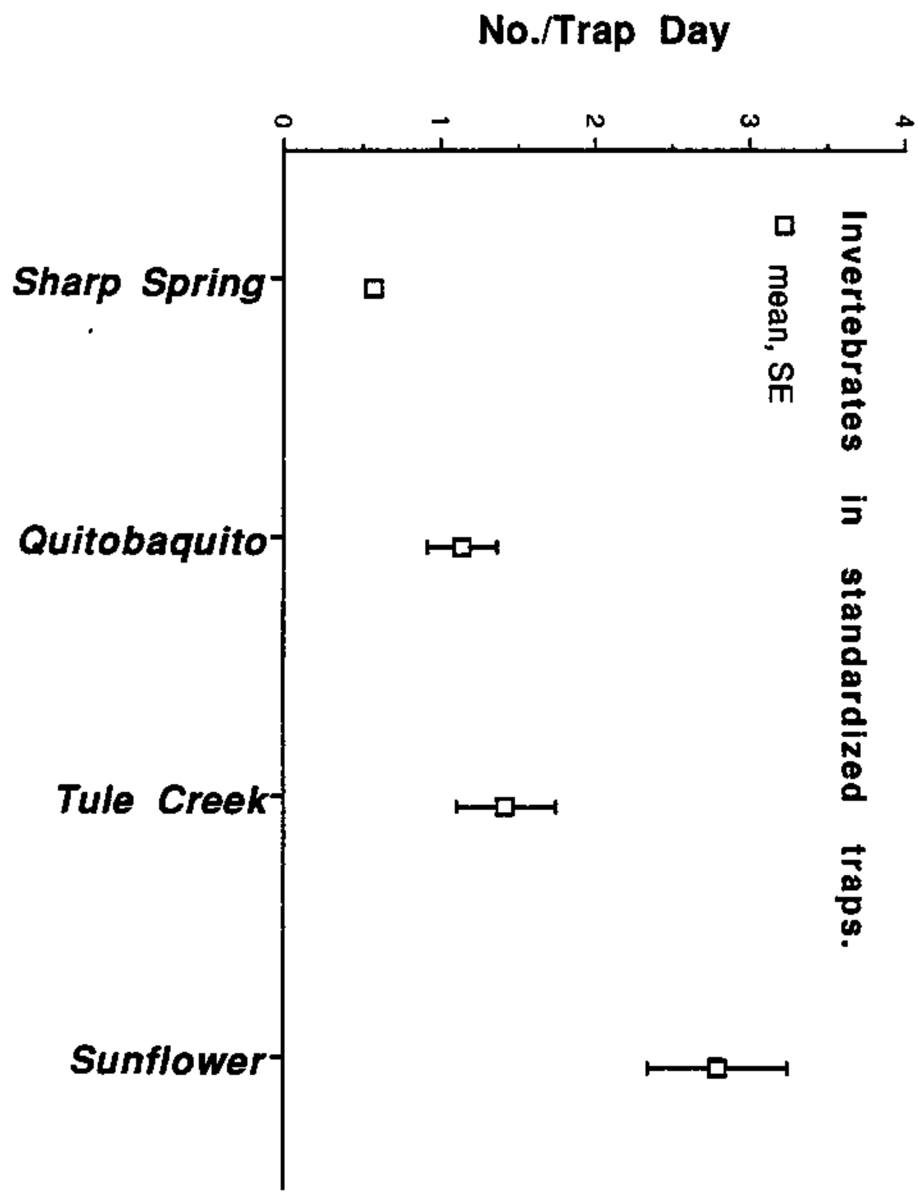


Figure 5. Turtle food availability index based on baited minnow traps set at water line. Entrances were opened to 5-6 cm.

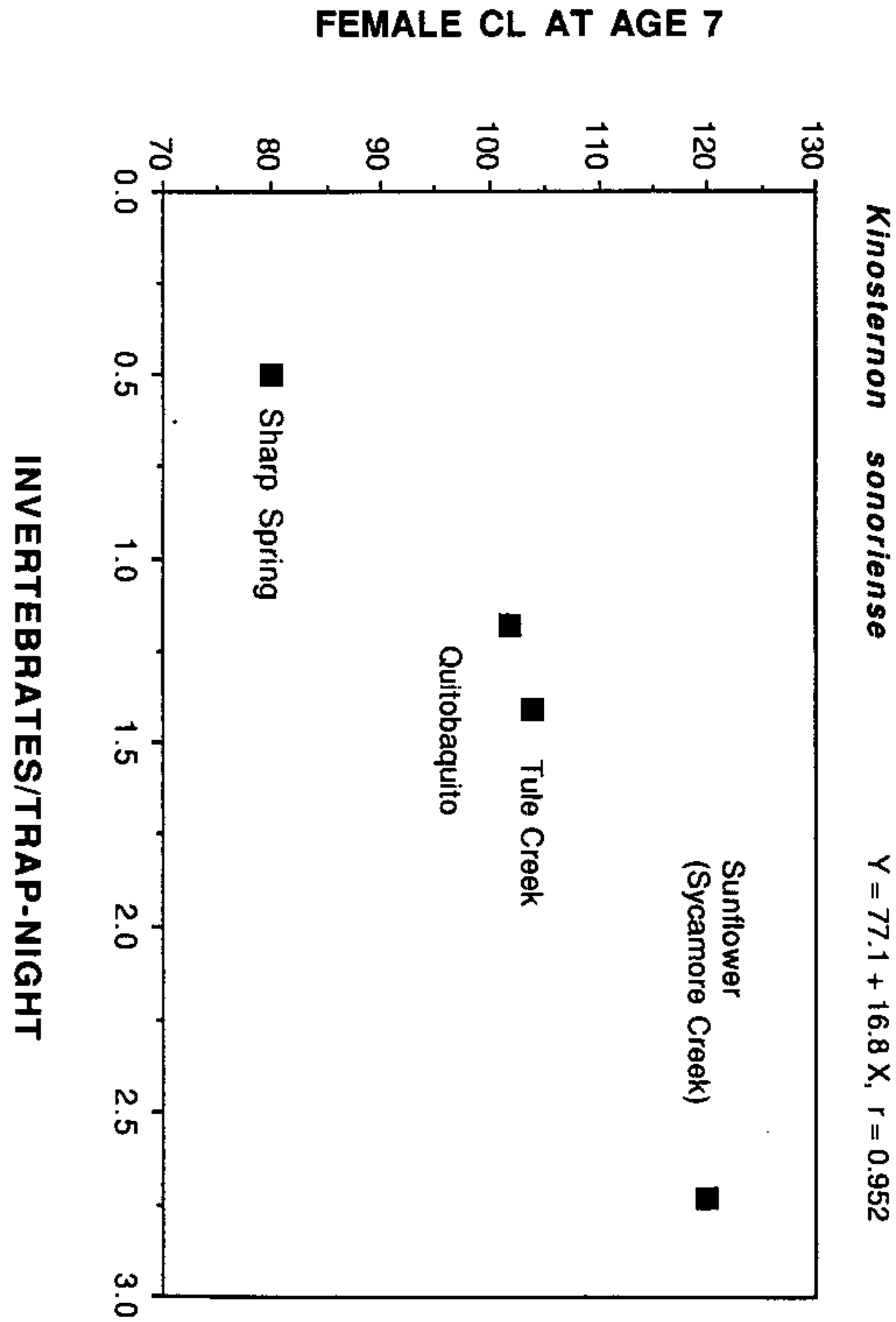


Figure 6. Relationship between individual growth and food availability among Sonoran mud turtle populations.

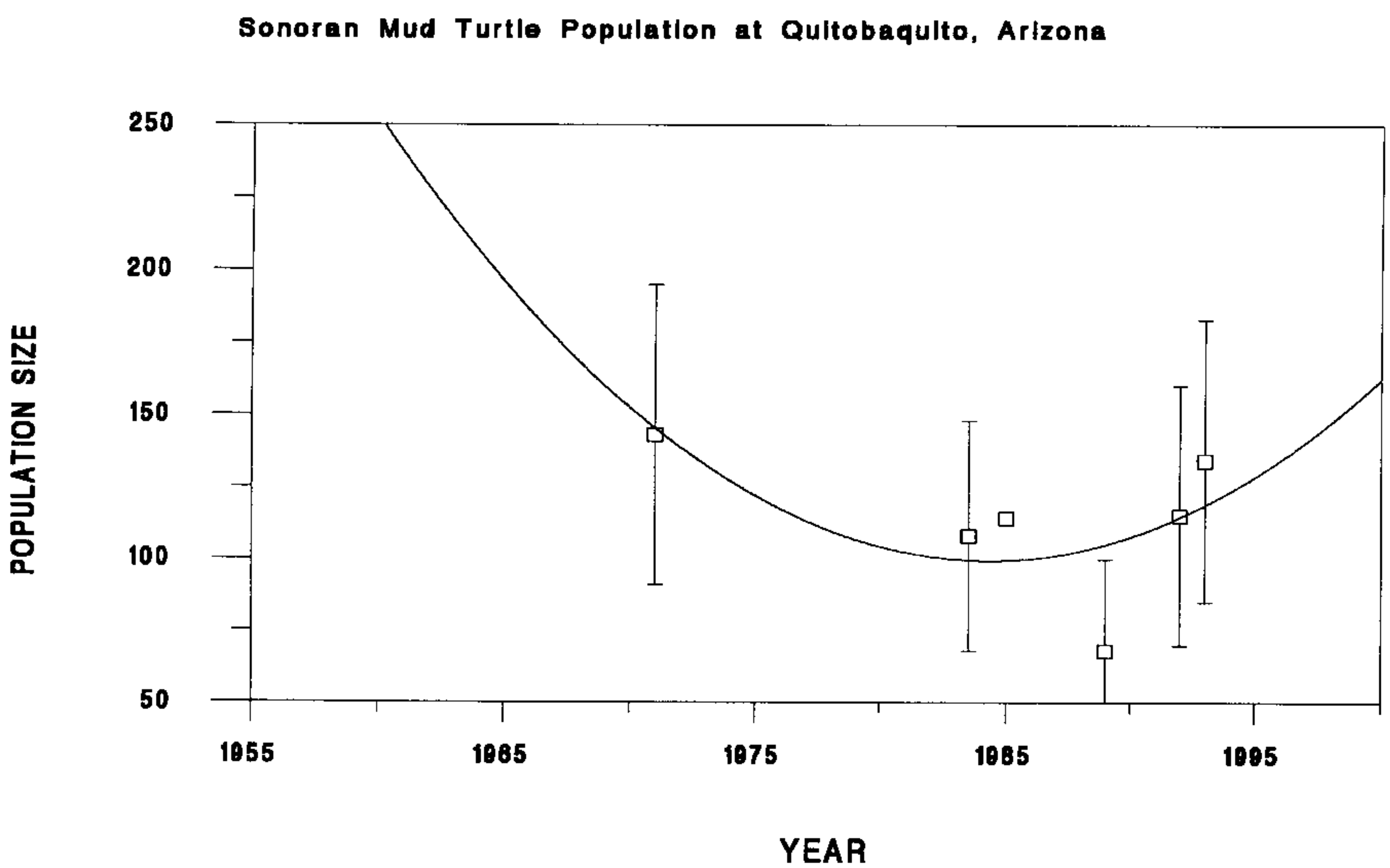


Figure 7. Fitted polynomial model of population trajectory based on data in Table 6.

DISCUSSION

Fourteen years of study of the Sonoran mud turtle at Quitobaquito have yielded an increasingly complete view of the life history and natural history of the population. We have substantial data to quantify key life history parameters, including age at sexual maturity, juvenile and adult survivorship, and individual growth and lipid storage. The difficulty of determining female reproductive frequency and clutch size in this small population is slowly being resolved. Patterns of recruitment, sex ratio, and population size are also becoming clear. The potential effects of chemical contamination and nutritional shortfall have been explored in greater detail than for other intensively studied turtle populations.

Several issues have arisen regarding the potential for survival of the turtle population at Quitobaquito, as well as concerning the effects of management in favor of the turtle population. First of all, was there ever a decline in numbers of mud turtles at Quitobaquito? Has this decline been reversed, and if so, how permanently? What demographic and environmental factors are most important in the issue of population decline or expansion? And hence, where, if anywhere, should research and management efforts be directed? The following discussion will attempt to treat each of these questions in turn.

Long Term Population Change Versus Stability

Fig. 7 shows the best available population estimates for the Sonoran mud turtle at Quitobaquito over approximately 30 yr. The impression of Scotty Steenbergh, a scientist and park naturalist at Organ Pipe for many years, was similar to Minckley's (personal communication) early impression of large numbers of turtles at Quitobaquito. Gehlbach's (1979) estimate at 143 seems to be consistent with the relatively high capture rates obtained independently by him (Gehlbach, 1979 and personal communication) and Hulse (1974a). The low population estimate for 1989 is below the computed 95% confidence intervals for 1971 (Gehlbach, 1979) and 1992 (this report).

There is evidence of medium-term fluctuations in recruitment into the juvenile and young adult age classes (Tables 8, 10), but evidence of substantial changes in the adult population is more tenuous. Clearly, when turtles were distributed as pets during one of the early pond management operations, the adult population was affected, but there is less direct evidence for changes in the adult population since 1970. Demographic data in the early 1980's are consistent with the apparent population minimum computed for the late 1980's, in two ways: (1) the juvenile population was confirmed to be relatively small at that time, and (2), the low recruitment pattern shown for the early-mid 1980's would be expected to translate into the observed decline in the adult population in the late 1980's.

In summary, there is moderately convincing evidence of a substantial population decline at Quitobaquito from 1960 or 1970 to the late 1980's. How much of the decline was systematic, rather than a product of normally varying recruitment in an extreme desert environment, remains open to question. Evidence for a rebound is also convincing, especially for successful recruitment of subadults in the 1990's. The question remains: is this a systematic rebound likely to be sustained and to yield substantially increased population density?

Life Table Synthesis

The current best estimate for the life table of the Sonoran mud turtle at Quitobaquito is in Table 12. This table uses the computed values for adult female and age-specific juvenile survivorship. Average annual clutch frequency is set at 1.5 (1.0 at ages 5 and 6), and clutch size increases from 3 (ages 5-8), to 4 (ages 9 and 10), and 5 subsequently. Sex ratio at birth is assumed to be 1.26 males per female, and fecundity adjusted accordingly. Net replacement rate R_0 = 1.57 indicates a substantially increasing population, as is consistent with population size computations. This result is auspicious for the population's future.

None of the estimates used in Table 12 are likely *a priori* to be biased toward high survival or reproductive rates. However, each variable estimate has a sizable associated error variance, and changes in any value will affect the outcome R_0. For example, if we use the mean rate of adult survivorship at 0.87, R_0 increases to 1.78, and similarly, assuming a 1:1 sex ratio at hatching yields $R_0 = 1.77$. Reducing the estimated clutch frequency to 1.0/yr (0.67/yr at ages 5-6) reduces R_0 to 1.05. By contrast, reducing survivorship at ages 3 and 4 to 0.62 (as at age 2), yields $R_0 = 0.99$; reducing these values to 0.50 yields $R_0 = 0.64$; and if s_x is set at 0.50 for ages 2-4 yr, R_0 falls to 0.52. Among the most reasonable variations we can impose on the estimates, parameter adjustments yielded a prediction of population growth ($R_0 > 1.00$); and several of the adjustments to juvenile survivorship led to the outcome $R_0 < 1.00$. The reduced juvenile survivorship values used apparently correspond to those actual occurring in the early-mid 1980's (Table 9; Rosen, 1987).

Although these results are consistent with the hypothesis that juvenile survivorship primarily affects population trajectory, we emphasize that our demographic estimation remains at a preliminary stage. A combination of two complementary changes in other demographic parameter estimates may also yield $R_0 < 1.00$. Increasing precision of all parameter estimates, but especially clutch size and frequency, would greatly increase our ability to rely on this kind of demographic analysis.

Causes of Population Change

For the following discussion, we assume that the estimated changes of the turtle population at Quitobaquito are real. The population was largest pre-1960, declined to about 140 by 1971, and about half that by 1989. By 1993, the population was back to about 130. Four hypotheses should be considered to account for these changes based on available information: (1) loss of juvenile habitat in the 1961 pond renovation reduced recruitment into the adult population, via increased predation on juveniles; (2) sex ratio anomalies drive major population fluctuations; (3) mortality associated with poor nutritional status drives the population down via attrition of adults; and (4) pesticide or metal toxicity based on natural or anthropogenic contamination of the environment leads to low survivorship. It is possible that all three processes are occurring. The question is, are any or all of them important?

Table 12. Life table for female Sonoran mud turtles at Quitobaquito Springs, Organ Pipe Cactus National Monument, Arizona, 1982-1995. This model assumes 1.5 clutch/yr, except in 6-7 yr olds, which average 1/yr. In addition, sex ratio at hatching is assumed to be 1.25 males:1 female.

Age (yr)	N_x	$1-q_x$	m_x	l_x	$l_x m_x$	$x l_x m_x$
0	60.0	0.900	0.00	1.0000	0.0000	0.00
1	54.0	0.280	0.00	0.9000	0.0000	0.00
2	15.1	0.620	0.00	0.2520	0.0000	0.00
3	9.4	0.730	0.00	0.1562	0.0000	0.00
4	6.8	0.840	0.00	0.1141	0.0000	0.00
5	5.7	0.855	1.33	0.0958	0.1277	0.64
6	4.9	0.855	1.33	0.0819	0.1092	0.66
7	4.2	0.855	2.00	0.0700	0.1401	0.98
8	3.6	0.855	2.00	0.0599	0.1198	0.96
9	3.1	0.855	2.67	0.0512	0.1365	1.23
10	2.6	0.855	2.67	0.0438	0.1167	1.17
11	2.2	0.855	3.33	0.0374	0.1248	1.37
12	1.9	0.855	3.33	0.0320	0.1067	1.28
13	1.6	0.855	3.33	0.0274	0.0912	1.19
14	1.4	0.855	3.33	0.0234	0.0780	1.09
15	1.2	0.855	3.33	0.0200	0.0667	1.00
16	1.0	0.855	3.33	0.0171	0.0570	0.91
17	0.9	0.855	3.33	0.0146	0.0487	0.83
18	0.8	0.855	3.33	0.0125	0.0417	0.75
19	0.6	0.855	3.33	0.0107	0.0356	0.68
20	0.5	0.855	3.33	0.0091	0.0305	0.61
21	0.5	0.855	3.33	0.0078	0.0260	0.55
22	0.4	0.855	3.33	0.0067	0.0223	0.49
23	0.3	0.855	3.33	0.0057	0.0190	0.44
24	0.3	0.855	3.33	0.0049	0.0163	0.39
25	0.3	0.855	3.33	0.0042	0.0139	0.35
26	0.2	0.855	3.33	0.0036	0.0119	0.31
27	0.2	0.855	3.33	0.0031	0.0102	0.27
28	0.2	0.855	3.33	0.0026	0.0087	0.24
29	0.1	0.855	3.33	0.0022	0.0074	0.22
30	0.1	0.855	3.33	0.0019	0.0064	0.19
				R_0 =	1.5730	
					T =	11.94

Hypothesis I. Predation on Juveniles. Four important lines of evidence support the hypothesis that juvenile survivorship is critical for population change at Quitobaquito. First, numerous predators are observed at Quitobaquito. A desert oasis may attract predators from a large surrounding area, concentrating their effects more strongly than a larger wetland. During 1984-1985, during 30 d on-site work, one of us (P. Rosen) observed 6 belted kingfishers, 6 great blue herons, 1 black-crowned night heron, 2 (great?) egrets, and 1 merganser, all foraging in the pond. The wading birds were often seen standing in shallow, open water at the edge of the tule mat (Fig. 2) where juvenile turtles were most often captured. It appeared that small turtles would be vulnerable if they emerged to forage from the protective cover of the emergent vegetation.

Second, juvenile and hatchling Sonoran mud turtles showed a clear preference for the kind of habitat--shallow, productive water with protective emergent vegetation--that was reduced by the digging operation in 1961, and was increased by the spring-channel operation in 1989-1990. Man-caused changes in juvenile habitat correlate well with what we know of population processes in this population.

Third, manipulations of the Quitobaquito population life table within the ranges of suspected parameter error variances indicate that changing juvenile survivorship results in dramatically shifting the population trajectory from growth to decline. Changing other parameters singly failed to show such a strong effect.

Finally, The increased number of surviving juveniles during the 1990's, following the reconstruction of juvenile habitat in the spring channel, suggests that management efforts to enhance juvenile survivorship may be succeeding. This is obviously the primary evidence of interest: is the population actually increasing? It apparently was rebounding, but given the temporal variation of recruitment at Quitobaquito in general, we cannot know yet whether the rebound is a temporary fluctuation, unrelated to management efforts, or a lasting recovery. Future sampling at Quitobaquito will help answer these questions. For the time being, we conclude that changing juvenile survivorship patterns appear to be most important for population dynamics of the Sonoran mud turtle at Quitobaquito.

Hypothesis II. Sex Ratio Anomalies. The occurrence of sex ratio anomalies in the Quitobaquito turtle population is suspected but not strongly demonstrated. The anomaly for the early-mid 1970's is statistically significant, but is dependent upon growth ring analyses of data collected up to 10 yr post-hatching, and hence is imprecise. The picture for the 1980's and 1990's is clouded by the difficulty of sexing juvenile females. The sex ratio anomaly observed--few females recruiting in the 1970's--could clearly contribute to the observed population trends. However, it cannot explain the change in juvenile survivorship observed between the 1980's and 1990's. Finally, if there is a global warming trend affecting Quitobaquito, this would be expected to lead to an increase of female hatchlings, which would, presumably, benefit the population, at least in the near term.

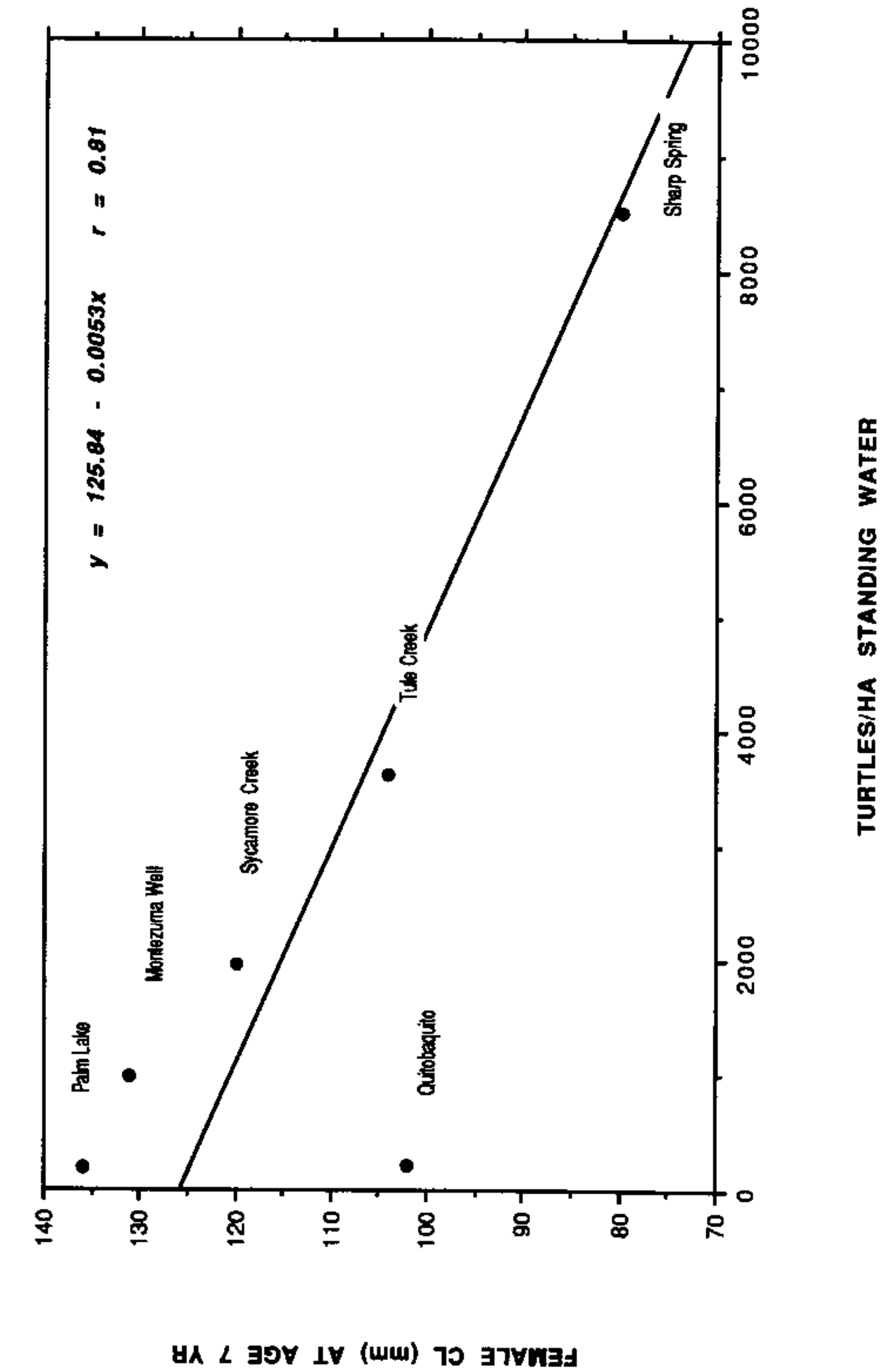
Hypothesis III. Nutritional Stress. The estimate of annual survivorship in adult females at Quitobaquito (85.5%) is lower than reported for a stable population of yellow mud turtles in Nebraska (*K. flavescens*, 95%, Iverson, 1991) and also lower than in an apparently declining population of common mud turtles in South Carolina (*K. subrubrum*, 87.6%, Frazer et al., 1991). It is also lower than in most other populations of the Sonoran mud turtle under study in Arizona (Rosen, 1987). While somewhat low, however, the observed rate of 85.5% is not off-scale, and may be small cause for concern if the observed population growth continues. It remains of scientific interest, and potential conservation importance, to identify the source of the unusual mortality.

It is evident, both from turtle growth rates and from aquatic invertebrate abundance, that food is in short supply for mud turtles at Quitobaquito. The nutritional stress or shortfall is probably also responsible for the relatively low clutch frequency (which may be as high as 4 clutches/yr in this species, compared to 1.5/yr at Quitobaquito [Rosen, 1987]). The low lipid storage rates found in the 8 analyzed carcasses also support the hypothesis that food is limiting. Furthermore, there was no evidence to support organochlorine or heavy metal toxicity in the turtles or in the habitat at Quitobaquito; and many of the carcasses showed no predation marks. Thus, the evidence suggests that poor nutrition, rather than a mysterious or anthropogenic disease, is the primary cause underlying the unusual mortality at Quitobaquito.

The lipid extraction data suggest that the turtles probably did not actually collapse from total depletion of energy-bearing fat stores. The turtle with the lowest reading (0.89% lipid wet-weight basis) was much lower than the others, and upon dissection it also was clearly the leanest. Unless the depleted substratum was a carbohydrate rather than a lipid, some secondary process probably contributed the direct cause of mortality. We did not detect signs of disease, although the condition of the carcasses was not fully conducive to autopsy. It may be that the turtles are in a weakened condition, and stress from any source may kill them via systemic physiological or behavioral failure.

Two leading limnologists in the Southwest have made observations at Quitobaquito. Cole and Whiteside (1965) reported substantial values for primary productivity, and Fisher (1989) explicitly noted that the aquatic ecosystem appeared normal, healthy, and productive. Despite this, Sonoran mud turtle growth at Quitobaquito was well below that expected based on studies in several other populations (Fig. 8; Rosen 1987). I hypothesize that competition with the thriving pupfish population plays a key role in trophic ecology at Quitobaquito.

Desert pupfish in general, and at Quitobaquito in particular, are well known to feed on many of the primary invertebrate groups eaten by Sonoran mud turtles (Schoenherr, 1988; Cox, 1972; Hulse, 1974b). Using a variety of methods, the pupfish population at Quitobaquito has been estimated at 1,408 - 8,907 in various years during 1975-1989 ($\bar{X}$ = 4780 $\pm$ 2,519 s.d., n = 12 years; summarized by Bagley et al., 1991). These pupfish densities (averaging nearly 15,000/ha in the 0.32 ha pond at Quitobaquito) are similar to or greater than the density used in an experimental pond study clearly demonstrating strong effects of desert pupfish on aquatic invertebrate abundance (Walters and Legner, 1980). Thus, competition with desert pupfish is the most plausible hypothesis to account for the several facets of nutrient deficiency in Sonoran mud turtles at Quitobaquito.



Kinosternon sonoriense populations in Arizona.

Hypothesis IV. Environmental Toxins. Although it is surprising that agricultural pesticides are found at such an isolated locality as Quitobaquito, the observed concentrations in the 1980's and 1990's in pond water and sediment, and in the turtle carcasses argue against pesticide intoxication as a source for the observed mortality or for population decline. Similarly, arsenic concentrations seem too low to have affected the turtles. On our data, we conclude that there is not strong evidence for a direct effect of chemical toxins on the mud turtles at Quitobaquito.

There are several key reasons that we should hold this conclusion reservedly: (1) turtles have not been tested directly for sensitivity to any of the potential chemical agents; (2) available studies in other ectothermic vertebrates are primarily for short-term, acute exposure (e.g., Fowler, 1983); (3) interaction of chemical exposures is possible in the turtles at Quitobaquito, and such effects could be synergistic, unpredictable, and poorly understood for any specific case (e.g., Fowler, 1983; McGeachy and Dixon, 1990; Peterle, 1991); (4) low organochlorine levels found in the turtles may reflect low lipid levels, where organochlorines may concentrate, rather than effectively low toxic levels; (5) agriculture and heavy use of such pesticides as parathion and DDT started in Mexico not far from Quitobaquito in about 1975, shortly before a major lag in turtle recruitment occurred (see above; Kynard, 1979, 1981; Brown, 1991), and (6) DDT and its metabolites, as well as m-parathion at 0.03 ppm were detected in Quitobaquito pupfish from 1976 and 1977 (Kynard, 1979, 1981; note, however, that while a population decline in pupfish was ascribed to m-parathion, no dead fish were reported, and levels observed may not necessarily reflect toxic exposure levels [e.g., Khan, 1977; Gasith and Perry, 1980; Hoffman et al., 1995]).

Hypothesis IV cannot be ruled out, despite our inability to develop supporting evidence. Environmental contamination remains a possibility that is potentially complex and difficult to demonstrate even with excellent study material (Don Norman, personal communication; see Appendix A), and hence one for which we should remain vigilant.

Directions for Further Monitoring and Research

The results of work to date indicate that the Sonoran mud turtle population at Quitobaquito declined from an early high in the 1950's to a low point in the mid-late 1980's, primarily resulting from degradation of juvenile habitat. Early results in the 1990's suggest that habitat refurbishment has improved juvenile survival and recruitment into the adult population. Our most immediate objective is therefore to re-census the population. We should determine (1) the full population size and (2) age structure and juvenile survivorship. The census will also assist in further refining estimates of adult survivorship.

Attention should be directed at the possibility that a male-biased sex-ratio anomaly is occurring in the Quitobaquito population. This kind of information will come in part from continued long term monitoring. Additional study of nesting in this or some other desert population of the species would assist an understanding of and modelling effort on this problem.

Regardless of population trends observed during monitoring, attention should be paid to any indication of unexplained vertebrate mortality at Quitobaquito. Turtles or pupfish found dead should be collected, stored on ice, necropsied, and frozen for chemical analysis. An indication of serious, increasing, or persistent unexplained mortality, especially if the pupfish became involved, would call for an intensive ecotoxicological study. Conversely, observations of natural mortality, such as by predation, are of great importance and relevance to understanding population processes.

RECOMMENDATIONS

Recommendations for conservation and management of the turtles at Quitobaquito include the following:

1. Recensus the population. Researchers, preferably the authors, should recensus the turtle population at Quitobaquito to obtain estimates for adult survivorship and current population size. This should occur at a minimum of 3 yr intervals to assure relatively accurate age-determination for unmarked juveniles.

2. Nesting study. A careful study of reproductive behavior is still lacking for this species. Use of small radio transmitters to track adult females to their nest sites should be done primarily at a different study area with more individual turtles.

3. Continue the protocol of collecting dead turtles. During the regular ORPI Resource Management monitoring and maintenance work at Quitobaquito, systematic searches of the pond, springs, and surrounding xeroriparian environment should be conducted. All dead turtles should be:

   (1) placed immediately on ice in a cooler
   (2) frozen in heavy duty ziplock freezer bags
   (3) identified with permanent-ink penned or with pencilled labels in the bag.
   (4) forwarded to research personnel at the University of Arizona for necropsy and storage.

4. Survey Río Sonoyta. Río Sonoyta should be surveyed to determine whether viable turtle populations persist there. A cooperative international venture, in which project personnel may train Mexican personnel in turtle study methods, would be appropriate.

ACKNOWLEDGEMENTS

There have been many major contributors to this project who are not listed as authors. Peter Bennett and Mike Kunzmann initiated the project in 1982, arranged with W.L. Minckley for the first author to assume the study in 1983, and have been a constant source of support for the work. Charles Conner, Mike Lee, Julie Parizek, and Robert McCord conducted much of the field work during 1989, 1992, and 1993, and Charles Conner, Ami Pate, and Tim Tibbitts collected the data in 1995. Ami Pate and Kirke King initiated the chemical analysis study of Quitobaquito sediments and turtle carcasses, which Kirke King and Cynthia Martinez then carried out, providing a major benefit to a project that lacked funding for such an analysis. Kirke King and John Moore, U.S. Fish and Wildlife Service at Patuxent, provided access to unpublished data on turtle lipid composition. Don Norman, Seattle, provided the assessment of requirements for a more thorough ecotoxicological study at Quitobaquito. Charles Conner and Ami Pate conducted regular surveys of the Quitobaquito environs, recovering turtle carcasses or skeletons and collecting data on live turtles observed or captured. Dr. Jim Jarchow, Tucson, examined live turtles in 1989. Tom Jones, Grand Canyon University, loaned the first author his canoe for the first three years of study, greatly facilitating the work, and Wendell Minckley at ASU loaned hoop nets to the project at that time. The old Ajo Hospital provided X-ray services in 1983, and Larry Nienabor at ASU assisted with oxytocin studies. The study could not have assumed anything like its final form without all of these collaborators.

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APPENDIX A

Toxicology Task Force

Avian Ecotoxicology and Biochemistry

Contaminants as a Cause of Death in Reptiles from a Sonoran Desert Oasis.

Donald Norman
March 1994

Summary

Concern about an apparent increase in observable mortality of turtles at a Sonoran Desert oasis has resulted in a summary below of possible causes of mortality from environmental contaminants. There are two likely sources of contaminants. The oasis waters and sediments may be altered by weather events, such as runoff from previous floods, leading to changes in bioavailability of contaminants. The second concern is about drift from pesticide use in the area. If drift is occurring, there may be danger to sensitive desert plants from herbicides and to some aquatic organisms from newer insecticides like synthetic pyrethroids. It is difficult to measure these newer pesticides in dead turtles and other information needs to be collected at the site. Unless evidence of organochlorine pesticides (OCs) can be documented in the area, or evidence of past heavy use, its role may be minimal.

This report was written to provide an overview of how to collect further information. It is written in a generic format because little information about the site were specified, and the immediate need was to assist biologists visiting the site to obtain that information. Photo documentation, collection, preservation, and storage of samples in the appropriate jars, and the ability to validate all aspects of the sampling event are necessary. A variety of inexpensive techniques are available to collect samples, but the procedures must often be adapted to field conditions depending upon the fate of certain suspected compounds. It is recommended that samples be collected, or information gathered about past sampling events at the site. Eco-epidemiology requires information from a wide range of geochemical, hydrological, atmopheric, cultural, agronomic, and industrial sources. This report was also written to demonstrate the need for a broad ecological basis for determining which species might be the best to demonstrate exposure or effects.

The Situation: Preliminary Diagnosis

Dead turtles were found at the Quitobaquito Oasis in Sonora in the Sonoita Valley south of Organ Pipe Cactus National Monument by Phil Rosen at the University of Arizona. The turtles found were quite emaciated. No necropsy was performed, but they were collected and frozen. Concern about possible impact of pesticide applications from nearby agricultural activities was expressed, but no direct water connections are present, limiting exposure to drift. A variety of crops are raised in the area, with cotton a major commodity. Historical use of organochlorine compounds in the area and dust from currently use insecticides present definate exposure routes to be investigated.

The Quitobaquito Oasis has no outlet, and with its high rates of evapotranspiration, is likely to accumulate any persistant compounds in its sediments. Creatures living in the sediments may have an elevated risk of exposure from a variety of sources such as dermal

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exposure and incidental sediment consumption. Mention of high levels of arsenic in the area raise the issue of elemental toxins as a source of mortality. Many of the the metalloids (arsenic, selenium, mercury) also have organometallic forms that are more toxic than their elemental or various oxidation states. Information about the transformation of these compounds has been elucidated, and areas in California with high levels of runoff, typically from agricultural areas, are experiencing mortality of amphibians. Exotic species may be more sensitive,

Possible Pesticide Exposure Scenarios and Methods of Interpretation

More information on local pesticides is needed. A list of pesticides provided (Table 1) could not be completely interpreted, as spanish names prevented identification of compounds. This list of pesticides used for 1987 may be irrelevant now. None of the persistant compounds now banned in the US were listed, but it is unlikely that they wouold have been listed. Because cotton, alfalfa, wheat, and fruit are raised in the area, just about any pesticide could be present. Therefore, information on current agricultural problems in the Sonoita Valley should be sought out by contacting local farmers and ranchers. The trend in much of Mexico and Central America is to switch from more persistent insecticides like toxaphene, DDT, and thiodan to more biodegradable organophosphate, carbamate, and pyrethroid insecticides like Sevin (carbaryl), parathion, and decis. While these newer insecticides are less persistent, they are much more acutely toxic, especially to aquatic organisms. Transport of these compounds is possible, and in certain types of aquatic systems, some of these compounds could persist, however, not as long as organchlorines. Degradation of many of these compounds is rapid in sunlight, and many are designed to be less toxic to vertebrates. It would take an extraordinary effort to document transport of pesticides, requiring the use of air samplers. Simple procedures require the collection of dust samples on filter paper, careful folding of the samples into vials, preservation with a solvent, and storage at freezing temperatures. If air pumps are available,

Pesticide Issues along the Borderlands: Old Ghosts that Don't Die.

Insecticides banned in the US decades ago are making a comeback. Not only are DDT, BHC, and other OCs still being produced in other countries, especially developing countries, resulting in the importation of these compounds on produce, but also new research has demonstrated that many of these compounds are more toxic than previously suspected. Historical studies have found elevated levels of DDT in many vertebrates that migrate across the border to winter in Mexico. While almost all insecticide use of organochlorines (DDT, chlordane, aldrin, toxaphene, endosulfan, dicofol) in the United States has been discontinued for a variety of reasons (legislative actions related to health, costs, and pest resistance), many third world countries can still obtain these insecticides and their low human toxicity makes them ideal in situations where people and not machines work the fields. Surveillance of imported crops from Mexico into the United States has reduced some use of these compounds, but the rates of contamination testing of imported produce has not kept up with the increase in imports.

Organophosphate Insecticides

The most toxic identifiable compounds on the provided list are organophosphates (OPs). OPs are more likley to be aerially sprayed, and therefore drift. Parathion is particularly toxic. Declines in insect populations might be apparent if the insecticide if they were being intensely studied. Aquatic effects might be more apparent if drift reached the water. Unfortunately, it is difficult to detect the OPs in tissue. The method of testing for exposure to OPs is measurement of cholinesterase inhibition, which requires brain or blood samples. The enzyme cholinesterase (ChE) is impaired from performing its function, and inhibition has a dose response. Measurement of ChE varies in different organisms, and may differ within the same ortganism depending upon the circumstances after the death of the organism. One solution would be to sample readily available organisms, such as fish, in the oasis, test for their ChE and subsequently determine if the level observed can be reactivated, which indicates an exposure to a ChE inhibitor. Typically samples are collected from brain, but work on plasma has also been successful. Many health departments will know of places to have samples analyzed for ChE inhibition. Fewer facilities will be able to perform the reactivation.

Analysis of Organchlorines

These classic insecticides are less likely to be of concern unless there is evidence that they are being illegally used. If there is evidence (empty cans) in the area, samples should be analyzed. Some dumping of insecticides has been reported. Methods are available to obtain a crude idea of the level of contamination. Because these compounds are lipid soluable and persistant, they biomagnify in fat tissue. An adipose sample can be taken through a series of preparatory steps to separate the compounds of concern from the fat to allow injection of a purified and concentration solution, to use the chemist's jargon, a series of extractions, cleanup, and concentration prepare a sample for gas chromatography, typically with electron capture detection (GC-ECD). It is important to perfrom the necropsy professionally, so that tissue weights and fat sampling is done without contamination. Many labs have their chromatographs calibrated to analyze for chlorinated compounds, and they can analyze the sample without getting fancy. Some labs are known to perform a few interesting samples for free, especially if there is potential for future work. One compound more difficult to analyze is dicofol, and it has been heavily used on cotton. OCs will be detected, they are universally present, so positive results do not necessarily indicate effects. Levels in the parts per million in lipid tissue would present a situation requiring more analysis. There may be interference from toxaphene, making analysis more difficult, and requiring an addtional separation phase.

Elemetal Toxicity and Water Quality Issues

Changes in water quality, from both droughts as well as El Nino rain storms can cause changes in the bioavailability of some toxic elements, and these changes might not occur immediately. Some elements, such as selenium, can be biotransformed. Selenium has become an important issue in many areas where irrigation occurs in seleniferous rock. Increased deposition of selenium in freshwater and marine sediments can in stimulate biotransformation of selenium into organo-selenium compounds 100 to greater than 1000 times more toxic than their form when they enter. Other such compounds include boron, arsenic, and mercury. The classic heavy metals, e.g. lead, cadmium, nickel, and copper also demonstrate variable toxicity with water quality changes. Changes in pH and alkalinity may cause eH-pH shifts in solubility of valence forms that vary in toxicity. Runoff may increase the concentration of elements in the water, so periods of high water may also have increased toxicity. Good information on standard water quality measures like alkalinity and pH, coupled with local geology can determine which compounds should be sampled.

Unfortunately, it is difficult to measure the organic forms of selenium, boron, arsenic, and mercury. The analysis requires different methods from the standard atomic absorption spectroscopy (AAS) used to measure most elements. New methods such as inductively coupled plasma spectroscopy (ICP) can measure many elements simultaneoulsy, providing a preliminary idea indication if selenium, boron, arsenic, and mercury are elevated. Samples of water and especially sediment should be collected on future trips. Sample jars must be cleaned to prevent traces of elements contaminating the sample, and frozen. This is particularly important for water samples. Sediments should be carefully collected to note when the sediment becomes anaerobic. Preferably a large number of samples should be collected, if there are various locations in the oasis that might have different sources of runoff.

Table 1. List of Pesticides used in Sonoita Valley in 1986-1987.

Trade Name	Chemical Name	Class	Analysis Method/Tissue
Tamaron	?	?	
Nuva Cron	?	?	
?	MZ330	?	
Sevin	carbaryl	organophosphate	cholinesterase inhibition stomach contents possible
-	parathion	organophosphate	cholinesterase inhibition stomach contents possible
-	decis	synthetic pyrethroid	detection very difficult aquatic insect mortality
Thiodan	endosulfan	organochlorine	would be present in fat, liver typically use GC-ECD for detection
Treflan	trifluralin	herbicide	Degrades rapidly, difficult to detect
Belmark	?	?	
Folimat	omethoate	organophosphate	cholinesterase inhibition, stomach contents possible

Conclusions ??

More questions than answers.

U.S. Fish and Wildlife Service

Region 2

Contaminants Program

CONTAMINANTS IN SONORAN MUD TURTLES FROM QUITOBAQUITO SPRINGS, ORGAN PIPE CACTUS NATIONAL MONUMENT, ARIZONA

by

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ABSTRACT: This opportunistic investigation explored levels and potential effects of organochlorine pesticides and heavy metals as factors limiting Sonoran mud turtles (*Kinosternon sonoriense*) at Quitobaquito Springs. Turtles found dead between 1989 and 1993 and pond sediments from Quitobaquito Springs were analyzed for pesticides and metals. Current levels of organochlorines are low and do not pose a threat to turtle survival and reproduction. Fat reserves in Sonoran mud turtles appeared relatively low suggesting an inadequate diet and possible dietary stress. Mean concentrations of boron, chromium, selenium, strontium, and zinc were significantly higher in turtles from Quitobaquito Springs than in softshell turtles from the highly contaminated Gila River. High concentrations of several elements in combination with a protein restricted diet may be a factor limiting turtle survival.

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The population of Sonoran mud turtles (*Kinosternon sonoriense*) at Quitobaquito Springs, Organ Pipe Cactus National Monument, Pima County, Arizona, has declined drastically since the 1950s when the turtles probably numbered in the hundreds (Rosen and Lowe 1996). By 1970, the population declined to about 143 individuals and by the early 1980s, Rosen and Lowe (1996) estimated that only about 100 individuals were present at Quitobaquito Springs. The reason for the decline was largely attributed to an inadequate food base, but organochlorine pesticides and heavy metals may also have played a role (Rosen and Lowe 1996).

Between 1989 and 1993, eight turtles were recovered dead from Quitobaquito Springs by the authors (PCR) and cooperators during radiotelemetry studies to determine survival, movements, and other population dynamics. An informal cooperative U.S. Fish and Wildlife Service/University of Arizona arrangement was completed in 1994 whereby the Service would chemically analyze the turtles found dead. This report summarizes organochlorine compound and heavy metal concentrations detected in the eight Sonoran mud turtles from Quitobaquito Springs.

METHODS

Turtles found dead were necropsied and results discussed by Rosen and Lowe (1996). After necropsy, turtle carcasses were stored frozen for chemical analysis. Whole body turtles were analyzed for organochlorine compounds and heavy metals.

To supplement the turtle analytical data, the authors (KAK and CTM) collected four sediment samples from Quitobaquito Springs October 12, 1994. Sediment samples were taken at relatively equidistant intervals along a north-south transect from the spring entrance to the pond's south shore. Sediment samples were weighed then placed on wet ice for about 12 hours before transfer to a commercial freezer. Sediments were analyzed for element content only (not pesticides).

Turtles were analyzed for organochlorine compounds and metalloids at Hazleton Environmental Services, Inc., Madison, Wisconsin. Samples were analyzed for p,p'-DDE, p,p'-DDD, p,p'-DDT, dieldrin, heptachlor epoxide, hexachlorobenzene (HCB), oxychlordane, *cis*-chlordane, *trans*-nonachlor, *cis*-nonachlor, endrin, toxaphene, mirex, and polychlorinated biphenyls (PCB). For each organochlorine analysis, the sample was homogenized and a portion mixed with anhydrous sodium sulfate and extracted with hexane in a Soxhlet apparatus for 7 hours. Lipids were removed by Florisil column chromatography (Cromartie et al. 1975). Sep-pak Florisil cartridges were used for removal of lipids (Clark et al. 1983). The organochlorine compounds were separated into four fractions on a SilicAR column to ensure the separation of dieldrin or endrin into an individual fraction (Kaiser et al. 1980). The individual fractions were analyzed with a gas-liquid chromatograph equipped with an electron-capture detector and a 1.5/1.95% SP-2250/SP-2401 column. Residues in 10% of the samples were confirmed by gas chromatography/mass spectrometry. The lower limit of quantification was 0.1 μg/g for all organochlorine pesticides and 0.5 μg/g for PCB. Organochlorine compounds are expressed in parts per million (ppm) wet weight.

Turtles and sediments were also analyzed for aluminum, antimony, arsenic, barium, beryllium, boron, cadmium, chromium, copper, iron, lead, manganese, mercury, molybdenum, nickel, selenium, strontium, vanadium, and zinc. Atomic absorption spectroscopy hydride generation was used to quantitate selenium and arsenic. Mercury concentrations were determined by cold vapor atomic absorption. All other elements were analyzed by inductively coupled plasma atomic emission spectroscopy. Blanks, duplicates, and spiked samples were used to maintain laboratory quality assurance and quality control (QA/QC). QA/QC was monitored by Patuxent Analytical Control Facility (PACF). Analytical methodology and reports met or exceeded PACF QA/QC standards. Element concentrations are reported in ppm dry weight. Mean concentrations of selected metals were compared between areas using a one-way Analysis of Variance (ANOVA).

RESULTS AND DISCUSSION

Organochlorines in turtles: DDE was the only organochlorine compound detected in turtles (Table 1). Residues were present in one-half of the samples and concentrations ranged from not detected to 0.035 ppm. Organochlorine residues at these levels are relatively low and probably would not have any adverse effects on turtle survival or reproduction.

Metals in sediment: Background data for most elements commonly occurring in Arizona soils are presented in Table 2. Unfortunately, no information is available for boron and strontium. Of the remaining elements, only selenium concentrations in Quitobaquito Springs sediments were above the range of normally encountered Arizona soils (mean ± 2SD). The threshold level of selenium in sediments above which effects on fish and wildlife might be expected is ≥4 ppm dry weight (Finley 1985, Garrett and Inman 1984). Selenium in Quitobaquito Springs sediments was 1.57 ppm or less, suggesting a low potential for selenium related problems.

Metals in turtles: Concentrations of seven elements designated by the Environmental Protection Agency (EPA) as priority pollutants were detected in turtles (Table 3). Apparently, ours is the first study to document contaminant levels of Sonoran mud turtles in Arizona, as we were unable to locate any data for meaningful within-species comparisons. To provide a frame of reference, we compared levels of elements in Sonoran mud turtle from Quitobaquito Springs with those detected in spiny softshell turtles (*Trionyx spiniferus*) collected from the lower Gila River (King unpub. data). Because of species differences in rates of pesticide and metal accumulation, this cross-species comparison is not entirely valid but, in the absence of Sonoran mud turtle comparative data, this cross-species comparison may aid in the interpretation of residue data.

Contamination of softshell turtles in the lower Gila River is well documented (Kepner 1987, Kepner unpub. data, King unpub. data); therefore, we expected that metal concentrations in spiny softshell turtles would be significantly higher than those in mud turtles collected from relatively pristine Quitobaquito Springs. However, this was not always the case. Only mean mercury concentrations were significantly ($P = 0.0014$, one-way ANOVA) higher in Gila River softshell turtles (0.362 ppm) than in Sonoran mud turtles (0.092 ppm) (Table 3). Mean arsenic and nickel residues were similar between species ($P \geq 0.1068$, one-way ANOVA). Mean concentrations of boron, chromium, selenium, strontium, and zinc were significantly higher in turtles from Quitobaquito Springs than in those from the Gila River ($P \leq 0.0012$, one-way ANOVA).

Arsenic: Background arsenic concentrations in biota are usually less than 1 ppm wet weight (3 - 4 ppm dry weight) (Eisler 1988). None of the turtle samples contained arsenic that approached this concern level.

Boron: Boron is a naturally occurring trace element generally considered environmentally innocuous, but boron has been documented to severely impair mallard (*Anas platyrhynchos*) reproduction at levels found naturally occurring in the environment (Smith and Anders 1989, Hoffman et al. 1990a). Elevated levels of boron are often associated with agricultural drainwaters. Hatching success and duckling survival was significantly reduced in feeding studies when mallard hens were fed 1000 ppm dry weight boron, less than one-third the highest boron concentrations found in plants of California's San Joaquin Valley. Additional research is needed to determine boron levels in aquatic plants at Quitobaquito Springs and assess their potential effects on Sonoran mud turtles.

Chromium: The organs and tissues of fish and wildlife that contain >4.0 ppm total chromium dry weight should be viewed as presumptive evidence of chromium contamination (Eisler 1986). Two of eight samples exceeded this concern level. However, we did not find any data that correlate concentrations of chromium in turtles with biological effects; therefore, even though two of eight samples contained elevated concentrations of chromium, toxicity cannot be established.

Copper: Copper is an essential dietary element for both plants and animals but at sufficient concentrations, copper may also be toxic (EPA 1980). Information is lacking on whole body residues and biological effects in many species, including turtles. We were unable to interpret the biological significance of copper at levels detected in the Sonoran mud turtles.

Mercury: Mercury concentrations are of special concern because mercury can bioconcentrate in organisms and biomagnify through the aquatic food chain. Mercury has no known biological function. The highest concentration of mercury detected in turtles from Quitobaquito Springs, 0.14 ppm dry weight, was below the 0.5 ppm level generally accepted as the concentration in biota from unpolluted environments (Abernathy and Cumbie 1977).

Selenium: Selenium is an essential trace element in animal diets, but it is toxic at concentrations only slightly above required dietary levels. No data were located regarding normal or background concentrations of selenium in turtles. Almost all, (18 of 19) softshell turtles from the lower Gila River contained selenium concentrations below the lowest level detected in Sonoran mud turtles from Quitobaquito Springs. The significance of selenium in the 1.63-2.11 ppm dry weight range as detected in turtles from Quitobaquito Springs is yet to be determined.

Strontium: The mean level of strontium in Sonoran mud turtles from Quitobaquito Springs was 4.8-times that in spiny softshell turtles from the Gila River (Table 3). No comparable data are available to assess whether strontium concentrations reported in this study were elevated or within the normal background range.

Zinc: Zinc is another essential element which at elevated concentrations can be toxic. Although tissue residues are not reliable indicators of zinc contamination, zinc poisoning usually occurs in birds and mammals when the liver or kidney contains >210 ppm dry weight. Zinc interacts with numerous other elements and the patterns of accumulation, metabolism, and toxicity from these interactions sometimes greatly differ from those produced by zinc alone (Eisler 1993). Mixtures of zinc/copper and zinc/nickel are generally acknowledged to be additive or more-than-additive in toxicity to a wide variety of aquatic organisms but, unfortunately, no data are available for turtles.

<u>Sources of contamination</u>: We are at a loss to explain why some elements appeared to be elevated in many Sonoran mud turtles. There are no obvious point sources of contamination in Quitobaquito Springs. Elevated levels may reflect aerial transport of contaminants, but additional research is needed to confirm this hypothesis. Since Quitobaquito Springs pond has no outlet during normal flow periods, the area may act as a sump for contaminants. Water is lost from the pond through evaporation; therefore, elements in the water column should tend to concentrate over time. We would expect that element levels in sediments to be especially elevated, but this was not the case.

<u>Reduced protein and metal interactions</u>: Reduced protein intake in combination with elevated metal levels may be suppressing turtle populations at Quitobaquito Springs. The Sonoran mud turtle is an omnivore that prefers invertebrates and fish when available (Hulse 1974). Aquatic invertebrates as food for mud turtles may be in chronic short supply at Quitobaquito Springs as evidenced by digestive tract and feces examination (Rosen and Lowe 1996). Body lipid reserves also were depleted in Sonoran mud turtles (mean = 2.81%) compared to those in softshell turtles (mean = 20.66%) indicating a possible dietary deficiency. In experimental studies with birds, protein deficient diets supplemented with elevated levels of arsenic, boron and selenium resulted in decreased growth rates and mortality (Hoffman et al. 1990b). Nutritional deficiencies may enhance certain element toxicity in birds and mammals and elements such as selenium can cause immunosuppression possibly rendering individuals more susceptible to disease (Hoffman et al. 1990b)

and stress. It is also possible that a protein deficient diet and high metal levels combined with the stress of capture and handling may have resulted in the unusual death of three of four radio transmitter-tagged turtles.

RECOMMENDATIONS AND SUGGESTIONS FOR FUTURE RESEARCH

This opportunistic investigation has raised more questions than it has answered. We offer the following suggestions on future research needs.

1. All turtles found dead should be salvaged and necropsied to determine which animal and plant species are being consumed. Carcasses should be stored frozen for further metal residue analysis.

2. Aquatic animals and plants most likely consumed by the Sonoran mud turtle should be collected and analyzed for selected trace metals to determine if environmentally hazardous levels are present.

3. Laboratory studies should be initiated to determine if current environmental levels of selected elements in combination with protein restrictions could be affecting adult Sonoran mud turtle physiology and reproductive success. Ideally, the test species should be the Sonoran mud turtle but, in the absence of populations large enough to withstand collections, a surrogate species such as the yellow mud turtle (*Kinosternon flavescens*) could be used.

4. Contaminant implications are obvious for the endangered desert pupfish (*Cyprinodon macularius*) which also occupies Quitobaquito Springs. Pupfish found dead should be salvaged for trace element residue analysis. Food items for pupfish should also be collected for residue analysis.

5. If contaminant concentrations are detected at high levels in pupfish samples, a surrogate species should be selected and laboratory studies initiated to determine levels of contaminants that could result in reduced survival and reproduction.

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