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Mary-Jane James-Pirri University of Rhode Island

Howard S. Ginsberg University of Rhode Island

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Authors

Mary-Jane James-Pirri, Howard S. Ginsberg, R. Michael Erwin, and Janith Taylor

Effects of Open Marsh Water Management on Numbers of Larval Salt Marsh Mosquitoes

MARY-JANE JAMES-PIRRI,¹ HOWARD S. GINSBERG,² R. MICHAEL ERWIN,³ AND JANITH TAYLOR⁴

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ABSTRACT Open marsh water management (OMWM) is a commonly used approach to manage salt marsh mosquitoes than can obviate the need for pesticide application and at the same time, partially restore natural functions of grid-ditched marshes. OMWM includes a variety of hydrologic manipulations, often tailored to the specific conditions on individual marshes, so the overall effectiveness of this approach is difficult to assess. Here, we report the results of controlled field trials to assess the effects of two approaches to OMWM on larval mosquito production at National Wildlife Refuges (NWR). A traditional OMWM approach, using pond construction and radial ditches was used at Edwin B. Forsythe NWR in New Jersey, and a ditch-plugging approach was used at Parker River NWR in Massachusetts. Mosquito larvae were sampled from randomly placed stations on paired treatment and control marshes at each refuge. The proportion of sampling stations that were wet declined after OMWM at the Forsythe site, but not at the Parker River site. The proportion of samples with larvae present and mean larval densities, declined significantly at the treatment sites on both refuges relative to the control marshes. Percentage of control for the 2 yr posttreatment, compared with the 2 yr pretreatment, was >90% at both treatment sites.

KEY WORDS open water marsh management, mosquito production, salt marsh

Grid ditching to control mosquitoes has been practiced on Atlantic coastal salt marshes since the early 1900s (Sebold 1992, Rozsa 1995, Casagrande 1997). By the late 1930s, \approx 90% of the Atlantic coast salt marshes from Maine to Virginia had been grid ditched (Bourn and Cottam 1950, Rozsa 1995). However, grid ditching negatively impacts salt marsh ecosystems by lowering water table levels, draining natural salt marsh pools, changing the natural vegetation community of the marsh, and decreasing the habitat value for a variety of wildlife (Bourn and Cottam 1950; Miller and Egler 1950; Daiber 1982, 1986; Clarke et al. 1984; Wolfe 1996; Adamowicz and Roman 2005).

A variety of manipulations, collectively referred to as open marsh water management (OMWM) (Ferrigno and Jobbins 1968), are currently used to provide relatively environmentally benign mosquito management, while lowering the use of pesticides to control salt marsh mosquitoes. OMWM involves hydrologic alteration to establish a salt marsh that is unsuitable for mosquito egg deposition and larval development, while promoting suitable habitat and access for larvivorous fishes (Ferrigno et al. 1975, Meredith et al. 1985, Wolfe 1996). The design of the hydrologic alteration varies regionally within Atlantic coast salt marshes, as each local mosquito agency tailors the design to specific marsh topography and current permit restrictions. For example, in the mid-Atlantic salt marshes (New Jersey and Delaware), a typical "traditional" OMWM design is used that includes ponds, radial ditches, and sills that partially retain tidal waters on the marsh; whereas in New England, ditch plugging, where plugs are installed in grid ditches allowing water to be retained in the ditches to create long rectangular pools, are common alterations. These approaches are now broadly used for the biological control and management of salt marsh mosquitoes, but controlled trials of the effectiveness of these approaches at lowering larval mosquito production are rare.

Here, we report the results of controlled field trials to assess the effects of two approaches to OMWM on larval mosquito production. These data were collected as part of a larger study that evaluated the ecosystem responses (e.g., vegetation, fish, and bird communities, water table level, and soil salinity) to these hydrologic alterations on salt marshes within U.S. Fish and Wildlife Service National Wildlife

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¹ Corresponding author: Graduate School of Oceanography, University of Rhode Island, Narragansett, RI 02882 (e-mail: mjjp@gso.uri.edu).

² USGS Patuxent Wildlife Research Center, University of Rhode Island, Kingston, RI 02881.

³ USGS Patuxent Wildlife Research Center, University of Virginia, Charlottesville, VA 22903.

⁴ US Fish and Wildlife Service, Region 5, Newington, NH 03801.

Refuges along the Atlantic coast (James-Pirri et al. 2008).

Materials and Methods

Site Selection. Study sites were salt marshes within Atlantic coast U.S. Fish and Wildlife Service National Wildlife Refuges (NWR). A Before, After, Control, Impact (BACI) study design (Stewart-Oaten et al. 1986) was established at Edwin B. Forsythe NWR (AT&T control and treatment sites), Oceanville, NJ (39° 41′ 47″ N; -74° 12′ 53″ W), and Parker River NWR (control and treatment site B2), Newburyport, MA (42° 46'43" N; -70° 48'33" W). At each refuge two historically paralleled grid-ditched marshes, one reference or control site and a corresponding treatment site that would be hydrologically altered, were chosen for study. Each control and treatment site were geographically close and experienced the same hydrologic regimes to minimize intrinsic marsh differences. At each location, both the control and treatment marsh were sampled for 2 yr before hydrologic alterations and for 2 yr after hydrologic alterations. In our BACI design, the hydrologic alteration (e.g., OMWM or ditch plugging) was the "impact" and the unaltered control marsh was the "control." All hydrologic alterations were conducted by local mosquito control organizations. More traditional OMWM-type practices were performed at Edwin B. Forsythe NWR, whereas ditch plugging with the creation of additional features such as deepening and sloping of the ditch edges (to facilitate bird usage), pond creation, and radial ditches occurred at Parker River NWR. The hydrologic manipulations were performed at the treatment sites in fall 2003 (Edwin B. Forsythe NWR) and in spring 2004 (Parker River NWR).

Mosquito Production. Mosquito production was evaluated by sampling mosquito larvae, using the standard dip count method, along randomly established transects that traversed the marsh elevation gradient from low marsh to high marsh. The first sample station was randomly located within the first 30-40m of the transect, and all subsequent stations were located at systematic intervals of 15-20 m depending on the site, yielding $\approx 40-60$ sampling stations within each marsh. This sampling design resulted in randomly located stations throughout each study marsh.

Larvae were sampled with a standard mosquito dipper (350 ml) 4 to 5 d after a tide that had flooded the surface of the marsh or 4 to 5 d after a major rainfall event, when salt marsh mosquito larval hatching was expected. Mosquito sampling stations were approached in the direction of the sun so that shadows would not be cast on the standing water and cause larvae to disperse. At each mosquito sampling station the nearest standing, stagnant water within a 3-m radius was located and sampled. All larvae were counted. To standardize the larval counts as an index of density (number per dipper), the amount of water present in the dipper was estimated using a scale from 0 to 5 (0, empty; 1, less than a one-fourth full; 2, one-fourth full; 3, half full, 4, three-fourths full; and 5, full). Density of larvae per dipper was then calculated using the following volumes on a 0-5 scale: 0, 0 ml; 1, 43.8 ml; 2, 87.5 ml; 3, 175 ml; 4, 262.5 ml; and 5, 350 ml. If no water was present then the station was recorded as "dry." A subsample of larvae from each sampling event was brought back to the laboratory for identification.

Mosquito data were analyzed using three different metrics: proportion of sampling stations that were wet (a proxy for potential mosquito production areas), proportion of sampling stations with mosquito larvae present (a proxy for potential mosquito production), and density of mosquito larvae (standardized by the amount of water in the dipper). Data for all dates within each year were averaged for each sampling station. Analyses of the proportion of sampling stations that were wet was performed on only those sampling stations that were wet at least once during the study (i.e., potential mosquito producing stations). Analyses of the proportion of sampling stations with larvae present and larval mosquito density were performed on only those sampling stations that produced larvae at least once during the study (i.e., mosquito producing stations) and were weighted by the number of wet sampling dates for each station in each year. Stations that were dry during the entire study period were omitted from analyses. Proportional data were arcsine transformed before analyses. Analyses of density data were performed on the ranked data. Full model repeated measures ANOVAs were performed, using the sampling station as the repeated variable for these parameters. A significant effect attributable to the hydrologic alteration was confirmed by finding a significant interaction term in the full model analysis of variance (ANOVA). When a significant interaction term was present, a least-squares means post hoc test was performed to determine which years, within each site, were significantly different from each other. The percentage of control was estimated using a modified Abbott's formula, where $100 \times [1 - ((\text{mean in treated area after treatment} \times$ mean in control before) / (mean in treated area before treatment \times mean in control after))] (Henderson and Tilton 1955).

On dates where numerous mosquito larvae were sampled the proportion of sampling stations with larvae present and the average density of larvae per 350-ml dipper were used to determine whether threshold criteria for the application of mosquito larvicide were approached or exceeded. Thresholds defined by the Delaware Mosquito Control Section (Delaware Mosquito Control Section 2008) were used as a guide to determine if dates when high abundances of mosquito larvae were sampled would have potentially triggered larvicide applications. The Delaware Mosquito Control Section uses the spatial distribution and density of mosquito larvae as indicators for possible larvicide application. Their thresholds are the presence of mosquito larvae in >25% of the sampled stations and an average larval density greater than five



Fig. 1. Influence of OMWM on mosquito production at Edwin B. Forsythe National Wildlife Refuge. (A) Proportion of mosquito sampling stations that were wet. (B) Proportion of samples with mosquito larvae present at mosquito producing stations. (C) Average larval mosquito density at mosquito producing stations. Summary of statistical comparisons among years, from least-squares means post hoc tests at P < 0.05.

larvae per dip (including wet dips with no larvae present or "zeros").

Results

Edwin B. Forsythe NWR, AT&T Sites. Proportion of Wet Sampling Stations. At the AT&T study sites, there were significant differences in the proportion of wet sampling stations (repeated measures ANOVA interaction term: F = 5.76; df = 4, 89; P = 0.0012). At the control site, the proportion of wet sampling stations was significantly higher in 2002 than in all other years (least-squares means, P < 0.05) (Fig. 1A). At the treatment site, differences in the proportion of wet sampling stations decreasing continually from 2002 to 2005 (Fig. 1A). Because the proportion of wet sampling stations was similar at the control from 2003 to 2005, whereas it steadily decreased at the treatment,

the decrease in wet sampling stations at AT&T treatment may be potentially attributed to the OMWM that occurred in fall 2003.

Presence of Larvae. Significant differences were observed at the AT&T sites in the proportion of sampling stations with mosquito larvae present (repeated measures ANOVA interaction term: F = 9.02; df = 3, 41: P = 0.0001). At AT&T control, differences were observed among all years except between 2002 and 2005 and between 2003 and 2004 (least-squares means, P <0.05) (Fig. 1B). At AT&T treatment, differences were observed between all years except between 2002 and 2003 (both years before OMWM) and between 2004 and 2005 (both years after OMWM) (least-squares means, P < 0.05) (Fig. 1B). Even though the control changed over time, the pattern of change was slightly different than that observed at the treatment site. At the AT&T treatment, the proportion of sampling stations where mosquitoes were present was significantly lower immediately after OMWM (in 2004) and then

Site	Date	Total no. wet stations sampled	% wet stations with larvae	Avg. larval density (no. per 350-ml dipper)	Avg. larval count (no. per dip)
Edwin B. Forsythe NWR					
AT&T control	12 Aug. 2002	40	18	6.3	5.7
AT&T control	4 Aug. 2003	16	56	132.3	42.4
AT&T control	5 Sept. 2003	42	38	7.5	3.6
AT&T Control	7 July 2004	32	34	12.3	5.6
AT&T control	17 Aug. 2004	39	46	10.3	2.5
AT&T control	19 May 2005	9	22	18	4.6
AT&T treatment (before OMWM)	15 July 2002	39	15	7.6	7.5
AT&T treatment (before OMWM)	12 Aug. 2002	46	15	3.3	1.1
AT&T treatment (before OMWM)	4 Aug. 2003	31	23	3.5	1.1
AT&T treatment (before OMWM)	5 Sept. 2003	39	10	10.3	1.8
Parker River NWR					
Control	25 June 2003	14	21	0.8	1
Control	18 July 2003	17	29	5.8	6.1
Control	15 Sept. 2003	11	55	16.3	5
Control	7 June 2004	32	16	3.3	4.6
Control	6 July 2004	34	41	8.3	9.2
Control	9 Aug. 2004	20	25	25.8	10.6
Control	12 May 2005	29	31	9.1	10.9
Control	27 June 2005	16	19	3.7	5.1
Treatment site B2 (before plugging)	25 June 2003	23	13	3.2	2.6
Treatment site B2 (before plugging)	17 July 2003	8	75	98.5	30.7

Table 1. Dates when larval mosquito production exceeded or approached larvicide threshold criteria used by the Delaware Mosquito Control Section

Dates when criteria were exceeded are indicated in bold. Edwin B. Forsythe NWR OMWM in spring 2004; Parker River NWR ditch plugging during 2004.

^a Delaware Mosquito Control Section threshold criteria for larvicide are the presence of mosquito larvae in >25% of the sampled sites and an average larval mosquito density of greater than five larvae per dip.

fell to zero in 2005, whereas at the AT&T control there was not a significant decrease in the proportion of sampling stations where mosquitoes were present between 2003 and 2004.

Larval Density. Mosquito larvae that were sampled at Edwin B. Forsythe NWR, AT&T sites included Aedes cantator Coguillett, Aedes sollicitans Walker, and Aedes taeniorhynchus Wiedemann. Significant differences were observed in mosquito larval densities at the AT&T sites (repeated measures ANOVA interaction term: F = 8.18; df = 3, 41; P = 0.0002). The same general pattern observed for the proportion of sampling stations with larvae present was also observed for larval density. At the AT&T control, significant differences were observed among all years except between 2002 and 2005, and between 2003 and 2004 (least-squares means, P < 0.05) (Fig. 1C). At the AT&T treatment, differences were observed among all years except between 2002 and 2003 (both years before OMWM) and between 2004 and 2005 (both years after OMWM), with higher larval densities observed in 2003 and 2004 (least-squares means, P <0.05) (Fig. 1C). Even though the control changed over time, the pattern of change was different than that observed at the treatment. At the AT&T treatment, larval mosquito density was higher before OMWM (2002 and 2003) and decreased to zero after OMWM in 2005, whereas at the AT&T control larval densities did not follow the pattern of lower densities in 2004 and 2005 than in 2002. Percentage of control comparing 2003 (before OMWM) to 2005 (after OMWM) was 100% (because there were no larvae on the treatment marsh in 2005). Comparing the two pretreatment with the two posttreatment years, percentage of control was 92.9%.

The control site at Edwin B. Forsythe NWR exceeded the larvicide criteria used by the Delaware Mosquito Control Section on two dates in 2003 and two dates in 2004 and approached (one of two criteria exceeded) the threshold on two other dates (Table 1). The treatment site never exceeded these criteria but approached the threshold on four occasions before OMWM alterations (Table 1).

Parker River NWR. Proportion of Wet Sampling Sta*tions*. There was a difference in the proportion of wet sampling stations at treatment site B2 among years (repeated measures ANOVA interaction term, F =5.69; df = 3, 94; P = 0.0013). At treatment site B2, the proportion of wet sampling stations was significantly different among all years except between 2002 (before ditch plugging) and 2006 (after ditch plugging) (leastsquares means, P < 0.05) (Fig. 2A). At Parker River control, differences in the proportion of wet sampling stations were observed among all years except between 2005 and 2006 (least-squares means, P < 0.09) (Fig. 2A), although changes in the proportion of wet sampling stations varied among years at the control, the pattern was similar between the control and treatment site B2, suggesting the number of wet sampling stations was not influenced by the ditch plugging at treatment site B2.

Presence of Larvae. There was a difference in the proportion of sampling stations with mosquito larvae present at mosquito-producing stations at treatment site B2 (repeated measures ANOVA interaction term: F = 14.07; df = 3, 30; P < 0.0001). At this site, the



Fig. 2. Influence of ditch plugging on mosquito production at Parker River National Wildlife Refuge. (A) Proportion of mosquito sampling stations that were wet. (B) Proportion of samples with mosquito larvae present at mosquito producing stations. (C) Average density of larval mosquitoes at mosquito producing stations. Summary of statistical comparisons among years, from least-squares means post hoc tests at P < 0.05.

proportion of sampling stations with larvae present was higher in 2003 (before ditch plugging) than in 2002 (before ditch plugging), 2005, and 2006 (2005 and 2006 were after ditch plugging) (least-squares means, P < 0.05) (Fig. 2B). At Parker River control, there were also differences among most years; the proportion of sampling stations with mosquito larvae present was significantly higher in 2004 than in all other years, and also higher in 2005 and 2003 than in 2002 (leastsquares means, P < 0.05) (Fig. 2B). Even though the control changed through time, the proportion of sampling stations with larvae were present was similar from 2003 to 2005 and in 2006, whereas at treatment site B2 the proportion of sampling stations with larvae present decreased from 2003 (before ditch plugging) to 2005 and 2006 (both years after ditch plugging). Therefore, it is likely that the decrease in the proportion of sampling stations with larvae present at treatment site B2 was related to the ditch plugging at this site.

Larval Density. Mosquito larvae that were sampled at Parker River NWR included Ae. cantator and Ae. sollicitans. At treatment site B2, there was a significant difference in larval mosquito densities (repeated measures ANOVA interaction term: F = 9.17; df = 3, 30; P = 0.0002), with higher densities observed in 2003 (before ditch plugging) than in 2002 (before ditch plugging), 2005, or 2006 (2005 and 2006 were after ditch plugging) (least-squares means, P < 0.05) (Fig. 2C). At Parker River control, larval densities increased from 2002 to 2004, when they were the highest, and then decreased from 2004 to 2005 and 2006 (Fig. 2C), and all years were significantly different except between 2002 and 2006, between 2003 and 2005, and between 2003 and 2004 (least-squares means, P <0.05). Even though the control site changed through time, the pattern of change was somewhat different from that observed at the treatment site. At Parker River control, densities were similar between 2003 and 2005, whereas at treatment site B2 they decreased

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from 2003 (before ditch plugging) to 2005 (after ditch plugging). Based on this pattern and that no larvae were sampled at treatment site B2 in 2005, the year immediately after ditch plugging, it is likely that the reduction in larval mosquito density was related to the ditch plugging at the treatment site. Percentage of control comparing 2003 (before ditch plugging) to 2005 (after ditch plugging) was 100% (because there were no larvae on the treatment marsh in 2005 (Fig. 2C). Comparing the two pretreatment with the two posttreatment years, percentage of control was 99.8%.

Parker River control exceeded the Delaware larvicide criteria on five dates, two dates each in 2003 and 2004 and one date in 2005; and approached these thresholds on three other dates (Table 1). At treatment site B2, the Delaware criteria were exceeded on one date and were approached on another date in 2003 (before ditch plugging) (Table 1).

Discussion

Our results provide evidence of effective reduction of mosquito larval densities on two salt marshes resulting from OMWM-related hydrologic alterations. On both marshes, larval densities declined in manipulated marshes relative to control marshes, resulting in a percentage of control of larval mosquito production in excess of 90% for at least 2 yr after the manipulations. Similar findings were reported for a recent study on Long Island, NY, where OMWM-type manipulations decreased the frequency of larval mosquito production by 70% on a treatment marsh compared with a control marsh (Rochlin et al. 2009). The effects of the interventions on the proportion of sample stations that were wet and on the proportion of sample stations with larvae were present shed light on the mechanisms of these two approaches to marsh management. The AT&T treatment marsh (traditional OMWM) exhibited declines in the proportion of mosquito sampling stations that were wet, but the Parker River treatment site B2 marsh (ditch plugging) did not. The manipulations at the AT&T treatment marsh included reexcavating existing ditches, creating new internal ditches and ponds, as well as ditch plugs to incorporate tidal ditches into a new closed pond and radial ditch system. At Parker River, the manipulations were closed tidal systems with ponds and radial ditches. The re-excavation and addition of new ditches presumably resulted in the decrease in the proportion of stations that were wet at AT&T treatment. The net drying effect at AT&T treatment also was reflected in a lowered ground water table level at this site after OMWM (James-Pirri et al. 2008). Thus, traditional OMWM practices seemed to use both draining of wet spots and creation of fish reservoirs as strategies to lower mosquito numbers, while ditch plugging relied more heavily on the fish reservoir approach. As mentioned, the control marshes at both of these sites also exhibited changes, but changes at the control sites had different patterns than those observed at the treatment marshes and, obviously, were not related to the hydrologic alterations at the treatment sites. Both

treatment marshes (at Edwin B. Forsythe NWR and at Parker River NWR), showed apparent decreases in the proportion of samples with mosquito larvae present at mosquito-producing stations and in larval densities after hydrologic alteration. Our results provide empirical corroboration of other studies that have observed decreases in mosquito abundance associated with OMWM and OMWM-type hydrologic alterations (Ferrigno and Jobbins 1968, Saveikis et al. 1983; Hruby et al. 1985, Daiber 1986, Lent et al. 1990, Wolfe 1992).

Delaware Mosquito Control Section larvicide application criteria were used as a guideline to determine if dates where high abundances of mosquito larvae were sampled would have triggered larvicide applications. These threshold criteria were exceeded at both control marshes, and on the treatment marshes before the hydrologic alterations, but not on the treatment marshes after the hydrologic alterations were performed. Therefore, these manipulations decreased the need for larvicide applications at these marshes. Rochlin et al. (2009) also observed a decreased need for larvicide applications after OMWM-type alterations on their treatment marsh. An additional note that is worth mentioning is that another treatment site at Parker River NWR (site A), which had undergone ditch plugging in 1994 to control mosquito production, also exceeded larvicide threshold criteria on isolated sampling dates during this study (James-Pirri et al. 2008). This underscores the necessity for continued larval monitoring even at sites that have historically been hydrologically altered.

The results from the Edwin B. Forsythe NWR sites were compromised somewhat by unexpected interventions. Larvicide (liquid Altosid, active ingredient methoprene, a juvenile hormone mimic) was applied to both the control and treatment marshes during the study period (in 2002 and 2004), possibly confounding the results. Altosid does not kill mosquito larvae directly but stops development and prevents adult emergence. Therefore, larval mosquito presence/absence data and density at the AT&T sites might not have been affected by the application of this larvicide. However, because the larvicide does prevent adult emergence it was possible that as the summer progressed the reduction in adult mosquitoes emerging from the marshes may have caused lower egg deposition on the larvicided marshes, which in turn could have resulted in fewer mosquito larvae on these marshes later in the season, thus confounding the results of the presence/absence and density data in subsequent years. Nevertheless, the effective mosquito control after OMWM in 2005 compared with before OMWM in 2003 at this site (both years after methoprene applications) argues for the effectiveness of the OMWM alterations.

We opted to use simple random selection of sample stations on the marshes to provide objective and repeatable results from the sampling program. More intensive sampling would certainly yield greater information on the changes in the spatial distribution of mosquito production (e.g., Rochlin et al. 2009) but may not be practical for routine monitoring of salt marsh mosquito production. Our choice of randomly located sampling stations results in a caveat in interpreting the results, because of the highly focal nature of larval distribution. Larval production at random sites on a marsh might not reflect trends at extraordinarily high-density sites that might be responsible for substantial proportions of the adult mosquitoes that later bite people. The fact that overall numbers of mosquito larvae declined in control as well as in treatment sites in 2005 exacerbates this concern. Therefore, a stratified random sampling program, with random sampling within high larval production sites. would be a logical subsequent study to confirm these results. Also, sampling of adult mosquitoes on treatment and control marshes before and after OMWM manipulations would provide more direct evidence on the effectiveness of this approach at lowering human exposure to mosquito bites.

Another difficulty is interpretation of results from the different nature of the hydrologic alterations at the two sites. Apparently, both traditional OMWM practices and ditch-plugging alterations can effectively lower mosquito larval numbers if applied appropriately; however, guidance for these alterations is general in nature and needs to be tailored to the specific topographic and hydrologic conditions on the marsh to be treated. Therefore, it cannot really be concluded from our results that "OMWM" effectively lowers mosquito larval production, but rather that the specific OMWM-type alterations performed on our study marshes lowered larval production at our study sites. Therefore, OMWM programs should follow an adaptive management approach, including monitoring to test effectiveness of the alterations, possibly leading to additional alterations in subsequent years to increase effectiveness or to mitigate undesirable outcomes, if needed.

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