U.S. Fish & Wildlife Service

The Aquatic Animal Drug Approval Partnership Program

"Working with our partners to conserve, protect and enhance the Nation's fishery resources by coordinating activities to obtain U.S. Food and Drug Administration approval for drugs, chemicals and therapeutants needed in aquaculture"

AADAP Aquatic Animal Drug Approval Partnership Program

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Makoshika State Park, Montana USA (Tom Bell)

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WHAT'S SHAKIN'

Authorization Granted for the Immediate Release of Fish Sedated with AQUI-S®20E under INAD 11-741

Based on a recent request, the U.S. Food and Drug Administration (FDA) has granted amended authorization for the use of AQUI-S®20E, a sedative drug, to allow for the immediate release of freshwater finfish sedated as part of field-based fisheries management activities. The amended authorization comes under the U.S. Fish and Wildlife Service's Aquatic Animal Drug Approval Partnership (USFWS-AADAP) Investigational New Animal Drug (INAD) 11-741.

Before the amended INAD authorization was granted, all freshwater fish sedated with AQUI-S®20E were required to be held for 72 hours—a withdrawal period impractical for field use. Although the immediate-release provision is for field use only and the withdrawal period remains at 72 hours for hatchery use, **rest assured this is some seriously good news!!**

FDA approval of an immediate-release sedative for use in fisheries management has been a high priority for the Association of Fish and Wildlife Agencies (AFWA), whose ongoing activities are coordinated by its Fisheries and Water Resources Policy (FWRP) Committee's Drug Approval Working Group. "This amended INAD authorization represents an enormous leap forward in our ability to effectively and safely sedate fish as part of field-based fishery management activities—activities that state and federal agencies and their partners use to restore, recover, protect, and manage fish populations that are important to the 48 million recreational anglers in the U.S., as well as to many others who depend on fish for sustenance and commerce," said Virgil Moore, Idaho Department of Fish and Game Director and chair of AFWA's FWRP Committee.

For more information about aquatic animal drugs, AQUI-S®20E, or to sign-up to participate in USFWS-AADAP INAD 11-741, go to *http://www.fws.gov/fisheries/aadap/home.htm* or contact the USFWS-AADAP INAD Administrator, Bonnie Johnson, at *bonnie_johnson@fws.gov* (phone: 406-994-9905). The full AFWA press release can be viewed at *http://www.fishwildlife.org/index.php?section=afwa_press_releases&prrid=180*.

Summary of the 18th Annual USFWS Aquaculture Drug Approval Coordination Workshop

The 18th Annual USFWS Aquaculture Drug Approval Coordination Workshop was held July 31, 2012, in La Crosse, Wisconsin USA, in conjunction with the

American Fisheries Society's (AFS) Fish Health Section annual meeting (August 1-3).

This year's workshop was hosted by the U.S. Geological Survey's Upper Midwest Environmental Sciences Center (UMESC), and Mark Gaikowski and other UMESC personnel (most notably Maren Tuttle-Lau and Jeff Meinertz) put together an excellent program. Technical sessions included (1) public agency and private sector overviews/perspectives on the current status of the aquaculture drug-approval process in the United States, (2) updates and status reports on aquaculture drugs currently being tested for initial or expanded label claims, and (3) an open-floor discussion of the day's presentations. At the conclusion of the technical sessions, the AFS Fish Culture Section Working Group on Aquaculture Drugs, Chemicals, and Biologics convened and updated workshop attendees on its recent and planned activities. Extracurricular sessions included two socials at local watering holes, a fun run along the Mississippi River, and an evening riverboat trip on the Mississippi River. Additionally, the AFS Fish Health Section meeting "kicked-off" with an aquaculture drug research and development technical session. Presentations given during this session included (1) an overview of the aquaculture drug approval process in the United States, (2) an overview of AADAP's National Investigational New Animal Drug (INAD) Program, and (3) information about how to publish efficacy and target animal safety data for consideration in aquaculture drug approvals.

Presentations from the Aquaculture Drug Approval Coordination Workshop can be accessed at *http://www.fws.gov/fisheries/aadap/inadworkshop12.htm*.

AADAP's Dr. Tom Bell Retires

Dr. Thomas A. Bell, who had worked in the U.S. Fish and Wildlife Service's (USFWS) AADAP Program since October 2003, retired at the end of May 2012. Tom was a huge part of the AADAP program, and although his primary/official role was New Animal Drug Application (NADA) coordinator, he was intimately involved in virtually all facets of day-to-day AADAP function. As NADA coordinator, Tom used his vast expertise and experience to help sponsors navigate through the U.S. aquaculture drug approval process. Tom was also the primary developer and administrator of our website, as well as our newsletter editor, MUMS grant proposal specialist, and special projects '"go-to" person. Tom also worked with AADAP's Bonnie Johnson and outside contractors to design and beta-test our new web-based INAD Program Management System (IPMS), as well as generate and manage the Public Aquaculture Fish Production Database.

One important—but underappreciated—contribution that Tom made to fish culture during his AADAP years was the derivation of a simple, generalized equation for

Dr. Tom Bell

correctly calculating either the (1) percent fish body weight to feed each day or (2) percent drug premix to incorporate into fish feed so as to administer a specific daily treatment dose (e.g., mg active ingredient per kg fish per day). This equation, based on prior—but unpublished—work by Bob Piper (USFWS, retired) and Jim Peterson (Montana Fish, Wildlife and Parks, retired), is now part of the USFWS Quick Desk Reference Guide to Approved Drugs for Use in Aquaculture (*http://www.fws.gov/fisheries/aadap/PDF/Flip-Book_FINAL%20for%20web%2023may2011l.pdf*, page 9-1).

Tom's career in aquaculture and fish health was long and productive. After graduating from Jamestown High School, Jamestown, New York, in 1966 and serving in the U.S. Navy (1968-1973), Tom earned his B.S. in Fisheries Science at the University of Washington (1978), his M.S. in Fish Health at Auburn University (1980), and his Ph.D. in Aquaculture and Chemotherapy at the University of Stirling, Institute of Aquaculture, Stirling, Scotland (1996). In addition to his AADAP years, Tom served as the USFWS National Fish Health Coordinator in Washington, DC (1999-2003); as a primary reviewer of applications for Investigational New Animal Drug (INAD) exemptions and New Animal Drug Approvals for nonmammalian aquatic species with the U.S. Food and Drug Administration Center for Veterinary Medicine in Rockville, Maryland (1992-1999); and as a researcher and teacher at the University of Arizona, Tucson (1980-1992).

Tom was senior author or coauthor of many peer-reviewed publications. Paramount among those was a book on the normal histology of Penaeid shrimp (Bell, T. A., and D. V. Lightner. 1988. *A Handbook of Normal Penaeid Shrimp Histology*. World Aquaculture Society,

Baton Rouge, Louisiana USA). This 120-page book was the first of its kind published for Penaeid shrimp and is still *the* book to consult on this subject.

Perhaps most important, Tom was (and remains) an excellent scientist—always curious and always thinking critically. When you want to "just talk science," you can always talk with Tom.

Although Tom (hb333slleb@gmail.com) has retired, he is staying in Bozeman and, in the near-term, will continue to assist with posting updates to the AADAP website. His NADA responsibilities have been temporarily absorbed by Dr. Dave Erdahl and Jim Bowker (i.e., takes two to replace one...right!!); Dan Carty has become the newsletter editor and MUMS grant proposal specialist; and Bonnie Johnson has assumed primary responsibility for administering the IPMS and Public Aquaculture Fish Production Database.

Tom—along with his science and curmudgeonly good cheer—will be truly missed. We wish Tom and his wife, Julie, all the best.

Text provided by Dan Carty (dan_carty@fws.gov), Fish Biologist, and Dr. Dave Erdahl (dave_erdahl@fws.gov), Branch Chief; USFWS AADAP; Bozeman, Montana USA.

American Fisheries Society Policy Statement on Fish Sedatives Kick-Starts Dialogue with Regulators, Including Discussion of Immediate-Release Options

Fisheries professionals in the United States have long needed legal access to a sedative where sedated fish can be immediately returned to the environment. Currently, the only legal option is to use a compound that requires sedated fish to be held for 21 d before they can be released and potentially captured for human consumption. The lengthy withdrawal period jeopardizes virtually every fisheries research project in which catchable-sized fish need to (or should) be sedated or anesthetized and then released into public waterways. This dilemma prompted the American Fisheries Society (AFS) Resource Policy Committee to draft a new policy statement on the need for an immediate-release anesthetic/sedative for use in the fisheries disciplines, calling attention to the need for better options for sedating fish during handling. In late 2011, the AFS adopted the new policy statement, and as part of a strategic plan to more effectively use policies statements to better inform others of AFS's position on specific topics, Dr. Gus Rhassam (AFS Executive Director) provided the approved policy to Drs. Bernadette Dunham (Director, U.S. Food and Drug Administration Center for Veterinary Medicine [FDA CVM]) and Steven Vaughn (Director, FDA CVM Office of New Animal Drug Evaluation [ONADE]), along with a request for a meeting of the AFS and FDA CVM "top brass" to discuss the content of the policy statement in greater detail. On April 25, 2012, leading representatives of AFS, FDA CVM, and the fish drug research and development community met for an unprecedented meeting at the FDA CVM offices in Rockville, Maryland USA. In an outstanding display of engagement, the leaders of virtually every FDA CVM office and team involved in fish drug approvals, including Drs. Dunham and Vaughn, came to the table for a frank discussion of sedatives issues. Many topics were discussed during this meeting, e.g.,

Why are compounds that are considered "Generally Recognized As Safe" in food considered risky if used to sedate fish that people will consume?

If quality control and manufacturing standards are tailored to the intended use in some areas of food and drug production, why are human drug manufacturing standards applied to fish drugs?

Given these purity, safety, and efficacy concerns regarding fish drugs, why are so many illegal products allowed to be directly marketed to fishermen and fisheries professionals?

The group plans to write an article for *Fisheries* magazine (an AFS publication), including FDA CVM's response to these and other "Frequently Asked Questions" regarding fish sedatives and other drugs.

Although the meeting represented a significant commitment of all stakeholders, one meeting—unprecedented or otherwise—cannot resolve all issues related to the fish drug approval process. As we hope to illustrate in the forthcoming 'FAQ with FDA CVM' article, standards are in place to ensure that approved and legally marketed drugs for use on fish are safe, effective, manufactured without impurities, and packaged and labeled according to FDA guidelines. In some instances, there is flexibility in how CVM approaches evaluations of drug efficacy and safety, and that mutually satisfactory strategies can be explored to make this process more efficient; in other situations, the way forward is less obvious. However, in taking an active role in setting fisheries standards and policies for our members, serving as an advisory resource in the development of fisheries-related public policy, and speaking for fisheries resources and fisheries professionals, AFS can assist our partners in increasing access to safe and effective fish sedatives and other drugs while protecting the public interest.

Text provided by Dr. Jesse Trushenski (saluski@siu.edu), Assistant Professor; Southern Illinois University at Carbondale; Fisheries and Illinois Aquaculture Center; Carbondale, Illinois USA and Jim

Bowker (jim_bowker@fws.gov), Research Program Manager; USFWS AADAP; Bozeman, Montana USA.

Information Quality in Regulatory Decision Making—the Discussion Continues

In the March 2012 issue of the AADAP newsletter, Dan Carty (AADAP) wrote an editorial about real and perceived differences between the academic and regulatory sciences and how those differences can affect aquaculture drug efficacy and target animal safety research in the United States. At the August 2012 meeting of the American Fisheries Society's Fish Health Section in La Crosse, Wisconsin USA, Dr. Susan Storey (FDA/CVM) gave an excellent presentation about publishing efficacy and target animal safety data for consideration in aquaculture drug approvals. Interestingly, many of the general issues raised by Dan and Dr. Storey have been—and continue to be—debated within the academic science and regulatory science communities. Of particular note is a review article by McCarty et al. (2012), which was recently published in the *Environmental Health Perspectives* journal. The article evaluates the rationale for regulatory decision making based on peer-review procedures versus Good Laboratory Practice (GLP) standards. The abstract is reprinted below with permission of the senior author.

McCarty, L.S., C.J. Borgert, and E.M. Mihaich. 2012. Information quality in regulatory decision making: peer review versus good laboratory practice. *Environmental Health Perspectives* **120(7):927-934**.

Abstract

Background: There is an ongoing discussion on the provenance of toxicity testing data regarding how best to ensure its validity and credibility. A central argument is whether journal peer-review procedures are superior to Good Laboratory Practice (GLP) standards employed for compliance with regulatory mandates.

Objective: We sought to evaluate the rationale for regulatory decision making based on peer-review procedures versus GLP standards.

Method: We examined pertinent published literature regarding how scientific data quality and validity are evaluated for peer review, GLP compliance, and development of regulations.

Discussion: Some contend that peer review is a coherent, consistent evaluative procedure providing quality control for experimental data generation, analysis, and reporting sufficient to reliably establish relative merit, whereas GLP is seen as merely a tracking process designed to thwart investigator corruption. This view is not supported by published analyses pointing to subjectivity and variability in peer-review processes. Although GLP is not designed to establish relative merit, it is an internationally accepted quality assurance, quality control method for documenting experimental conduct and data.

Conclusions: Neither process is completely sufficient for establishing relative scientific soundness. However, changes occurring both in peer-review processes and in regulatory guidance resulting in clearer, more transparent communication of scientific information point to an emerging convergence in ensuring information quality. The solution to determining relative merit lies in developing a well-documented, generally accepted weight-of-evidence scheme to evaluate both peer-reviewed and GLP information used in regulatory decision making where both merit and specific relevance inform the process.

Environmental Health Perspectives is an open access journal; hence, the full article can be downloaded at *http://ehp03.niehs.nih.gov/article/fetchArticle.action?articleURI=info%3Adoi%2F10.1289%2Fehp.1104277*.

AADAP DRUG UPDATES

General—Summer is flying by, the threat of wildfire is omnipresent due to drought conditions, not a single effectiveness trial has been conducted in the past few months, but we're hip-deep in conducting target animal safety (TAS) studies. The first two remarks are not too unusual this time of year, but the last two are very unusual. Since our involvement in conducting studies to support aquaculture drug approvals, summertime has typically been the time we dedicate to conducting pivotal field effectiveness trials and planning TAS studies to be conducting during the winter months. This year, however, we're getting ready to launch our 3rd consecutive TAS study with one additional study waiting in the wings. As an added bonus, two of the studies have been conducted in our all-new redesigned, retrofitted, and generally "modernized" AADAP Drug Research wet-laboratory. The wet-lab still has that "new car smell," and we all found it rather enjoyable working in an area better suited for our needs. Somebody famous once said "if you build it, they will come." We've built it (or at least modernized it), now we're putting the wet-lab to good use. For an update of our research activities, read on!

AQUI-S®20E (10% eugenol) and BENZOAK® (20% benzocaine)

A whole bunch of efficacy studies—In the last AADAP Newsletter, we described the pivotal and high quality supportive effectiveness studies that we conducted during summer/fall 2011 at a number of locations around the country—e.g., AADAP wet-lab; Southern Illinois University, Carbondale, Illinois USA; U.S. Geological Survey's Upper Midwest Environmental Sciences Center, La Crosse, Wisconsin USA; Iowa

Department of Natural Resource's Rathbun Fish Culture Research Facility, Moravia, Iowa USA; and Alaska Department of Fish and Game's Fort Richardson Fish Hatchery, Fort Richardson, Alaska, USA. All the numbers have been crunched, and it turns out we did a whole lot more than originally required by CVM to demonstrate that each drug was an effective fish sedative. We conducted 20 studies to demonstrate that AQUI-S®20E (10% eugenol) effectively sedated 13 different fish species to handleable. Results from these studies were written up in 16 Final Study Reports (FSRs) that were all submitted to CVM by May 3, 2012. We conducted 17 studies to demonstrate that BENZOAK® (20% benzocaine) effectively sedated 12 different fish species to handleable. Results from these studies were written up in 12 FSRs that were all submitted to CVM by April 10, 2012. In a nutshell: salmonids (rainbow, cutthroat, lake, and brown trout) were sedated with 25 mg/L eugenol or 40 mg/L benzocaine at a water temperature of about 14°C; coolwater fish (yellow perch, walleye, common carp, and fathead minnow) were sedated with 40 mg/L eugenol or 80 mg/L benzocaine at a water temperature of about 18° C; and warmwater fish (sunshine bass, blue and channel catfish, and tilapia) were sedated with 60 mg/L eugenol or 150 mg/L benzocaine at a water temperature of about 24°C. Based on results from the pivotal studies, mean time for salmonids, coolwater fish, and warmwater fish to become handleable with (1) AQUI-S®20E was 1.8 min, 2.5 min, and 1.3 min, respectively and (2) BENZOAK® was 1.8 min, 2.1 min, and 1.2 min, respectively. Mean times for all fish to recover from sedation, regardless of sedative used, ranged from 3.8 to 9.9 min.

At this time, we have not yet submitted a letter to CVM requesting that the effectiveness technical section be considered complete for each sedative "...for sedation to handleable in all freshwater finfish." The game-plan is to incorporate the information that CVM provides us in their response letters to our request for formal review of each FSR to draft the technical section complete request letters. We anticipate that the effectiveness technical section complete letters will be submitted to CVM by the end of October 2012.

Target animal safety (TAS) studies—After we got the effectiveness studies out of the way, our focus shifted to completing three TAS studies to demonstrate there is an adequate margin of safety associated with sedating rainbow trout, yellow perch, and channel catfish to handleable with AQUI-S®20E. There are many difficulties associated with conducting safety studies on sedatives, including: (1) determining the highest proposed efficacious dose and overdose dose, (2) coming to agreement with CVM on the definition of an "adequate margin of safety" for a sedative at the highest efficacious dose and overdose dose (is it 2 min or 10 min or…), and (3) identifying exposure durations that demonstrate how long fish can be overexposed with 100% survival, as well as the breakpoint where the survival level becomes unacceptable.

When we finally settled on defining the term "margin of safety" for the highest efficacious dose and overdose dose, it was time to sedate lots of fish to determine what dose/duration combinations to test. Starting with rainbow trout, we determined that 40 mg/L eugenol was going to be the highest efficacious dose where fish could be overexposed for 3-4 min with acceptable survival (≥98% survival), and 60 mg/L eugenol was going to be the overdose dose where fish could be overexposed for 2-3 min with acceptable survival. After these parameters were established, conducting the study was relatively straightforward, and the in-life phase of the study was completed on June 28, 2012. Preliminary results showed that under the test conditions (15°C), fingerling rainbow trout could be overexposed with (1) 40 mg/L eugenol for 3.5 min and 5.5 min with high survival (98% and 96%, respectively), and (2) 60 mg/L for 2.1 min and 2.9 min with high survival (98% and 96%, respectively). We've just begun to process 10-fish samples from each replicate for histological evaluation, and we hope to get the report from our pathologist (Beth MaConnell; Headwaters Fish Pathology, LLC) by late fall/early winter. Based on extensive preliminary testing in which we overdosed and overexposed rainbow trout with isoeugenol and eugenol, we do not expect any test-article related lesions of concern to be detected in the evaluated tissues.

Next on the TAS list were plans to conduct a similar study to evaluate the safety of the highest proposed efficacious dose and overdose dose on fingerling yellow perch. We received 2,500 test fish on July 30, 2012, from Pleasant Valley Fish Farm (McCook, Nebraska USA) and, during early August, conducted preliminary testing to identify appropriate dose/duration combinations for testing. The in-life phase of the study was launched on August 22, 2012, and should be complete by the end of the month. Although the initial data "look good," we will wait until the next Newsletter issue to provide a more complete update on study results.

In an effort to keep the TAS-ball rolling, we are planning to receive fingerling channel catfish from Pleasant Valley Fish Farm sometime in mid-September 2012 and conduct the final warmwater AQUI-S®20E TAS study shortly thereafter. We're really moving along with these TAS studies, and it's our plan to have all the AQUI-S®20E safety study FSRs submitted to CVM by the end of the year.

Channel Catfish Pituitary

Environmental assessment update—Based on a new round of comments from CVM's Environmental Safety Team (EST), a revised channel catfish pituitary Environmental Assessment (EA) was developed and

submitted to CVM on March 16, 2012. We are cautiously optimistic that the revised EA will be accepted by the EST because they provided clear and concise instructions relative to the information they wanted included in the EA. As an added bonus, I met with Drs. Holly Zahner and Eric Silberhorn and informally discussed each CVM comment and how the comment had been addressed. I left that meeting with the impression that the comments had been addressed reasonably well and that, although the EA may require some minor tweaking, it should otherwise be considered acceptable. We won't hear back from CVM's EST until sometime in November, so stay tuned.

In the meantime, we have been working with Drs. Chris Green (Assistant Professor of Aquaculture, Louisiana State University, Aquaculture Research Station) and Pat Gaunt (Mississippi State University, College of Veterinary Medicine) to develop protocols to evaluate the effectiveness and safety of peptides (e.g., channel catfish pituitary) as a spawning aid. Also, we've engaged with CVM Biotherapeutics Team to discuss completion of the Chemical, Manufacturing, and Controls data requirements for this crude product. Although the finish line for obtaining an approval for this product remains distant, we're making progress!

SLICE® (0.2% emamectin benzoate)

Efficacy studies on *Salmincola californiensis*—In the last issue of the AADAP Newsletter, we reported that CVM had accepted results from three SLICE® effectiveness studies we conducted to control infestations of *S. californiensis* in rainbow trout. CVM stated that results demonstrated "substantial evidence" of effectiveness. We also reported that although CVM could not provide to us an Effectiveness technical section complete letter for the use of SLICE® for this claim, a fourth effectiveness study had been conducted at the Missouri Department of Conservation's Maramec Spring Hatchery (St. James, Missouri USA). Well...we're happy to report that the fourth study was the ticket and that we received a letter from CVM stating, in part, that: "Based upon the information you submitted on October 21, 2011, and December 8, 2011, and the information contained in the investigational new animal drug file (INAD 11370), we consider the Effectiveness technical section to be complete. The technical section is complete for the use of emamectin medicated feed for the control of *Salmincola californiensis* in freshwater-reared *Oncorhynchus mykiss*." This was very exciting news because it puts us (and the drug sponsor) one step closer to completing a new animal drug application package.

Target animal safety study underway—With the effectiveness data requirements taken care of and out of the way, our focus on SLICE® shifted to completing the target animal safety technical section. As mentioned in the last issue of the AADAP Newsletter, we reported that we were preparing to launch the in-life phase of a study to demonstrate the safety of emamectin benzoate orally administered in feed to fingerling rainbow trout. During the in-life phase of the study, which was conducted March 20-April 3, 2012, fish were fed SLICE® -medicated feed at 0×, 1×, 2×, or 3× the proposed efficacious dose (50 µg emamectin benzoate per kg fish body weight per day) for 14 d (2× the proposed efficacious duration of 7 d). Preliminary results showed that (1) there was no mortality during the study, (2) fish behavior was characterized as normal, (3a) fish in the 0×, 1×, and 2× exposure groups consumed all feed offered during each feeding event ~95% of the time and (3b) fish in the 3× exposure group consumed all feed offered during each feeding event ~75% of the time, and (4) no gross lesions were detected in fish sacrificed postexposure for histological evaluation. Samples processed for histological evaluation are currently being examined microscopically by histopathologist Beth MacConnell, and we anticipate getting a pathology report from her relatively soon.

Previously conducted emamectin safety/tolerance studies on Atlantic salmon and rainbow trout at doses higher than those used in our study showed that the only signs of toxicity were behavioral (e.g., inappetance, dark coloration, loss of equilibrium). Based on this information, we anticipate histological findings to be unremarkable without detection of test-article related lesions. However, you never know what you're going to get until you get it. So, stay tuned.

35% PEROX-AID® (35% hydrogen peroxide)

Possible expansion of new label claim—We often use the phrase "bits and pieces" when working towards approval of a new drug or expanding an existing label claim. This phrase seems appropriate for our collective efforts to try to expand the 35% PEROX-AID® claim to include use to control infestations of *Gyrodactylus* spp. on freshwater-reared salmonids. Sufficient data have been submitted to and accepted by CVM that demonstrate hydrogen peroxide effectively controls or reduces infestations of *G. salmonis* in Coaster brook trout (study conducted by USGS UMESC and USFWS Genoa NFH staff – Genoa, Wisconsin USA) and rainbow trout (study conducted by USFWS AADAP and Ennis NFH staff – Ennis, Montana USA).

Concern has been expressed by the fish culture/health community that a label restricted to just *G. salmonis* would be problematic, mostly because very few, if any, fisheries professionals identify these critters down to species. Therefore, it was proposed to CVM that the label should allow use of 35% PEROX-AID® for use to control infestations of *Gyrodactylus* spp. native to North America (NA). CVM responded by stating that justification (e,g., published literature, non-USA data, and supportive studies) need to be submitted to support such a claim.

In response to CVM's request, Thomas A. Bell (AADAP-retiree) prepared a "white-paper argument" to support the proposed use of successful pivotal effectiveness studies conducted on *G. salmonis* to be considered representative of expected results on all species of North American *Gyrodactylus*. Information in the white paper included (1) a list of salmonid species native to NA and their associated parasitizing *Gyrodactylus* spp. isolated in NA; (2) a list of *Gyrodactylus* spp. infesting salmonids native to NA, but not isolated in NA (e.g., isolated in Europe); (3) chemical efficacy studies on *Gyrodactylus* spp. or other monogeans reported in the literature; (4) biological rational for similarity of treatment responses among *Gyrodactylus* spp. based on (a) basic unifying characteristics of the genus *Gyrodactylus*, (b) tegumental characteristics, (c) the limited number of *Gyrodactylus* spp. in NA (based on survey results and definitive diagnosis by Eric Leis – USFWS La Crosse Fish Health Center – La Crosse, Wisconsin USA), and (d) drug mode of action and why it should not differ between species. In conclusion, the document stated that the "prospective claim to be added to the existing 35% PEROX-AID® label should be for the control of *Gyrodactylus* spp. on freshwater-reared salmonids." We've had mixed success relative to providing justification for expanding a proposed label rather than providing results from pivotal effectiveness trials, so stay tuned.

Editor's Good News Flash—CVM recently notified us in writing that they consider the Effectiveness technical section to be complete "...for the use of 35% PEROX-AID (hydrogen peroxide) for the treatment and control of *Gyrodactylus* spp. in freshwater–reared salmonids when administered at 50 mg/L for 60 minutes or 100 mg/L for 30 minutes once per day on alternate days for three treatments in a continuous flow water supply or as a static bath." In their notification, CVM commented that the justification for this genus-level claim is based on information that demonstrates that *G. salmonis* is the most prevalent and pathogenic species of *Gyrodactylus* infesting freshwater-reared salmonids in the USA, and subsequently, that the effectiveness data submitted for *G. salmonis* infestations are representative of an overwhelming majority of the expected use pattern for an approved product. Also, CVM commented that the nonspecific nature of hydrogen peroxide treatment, physiological similarities among gyrodactylids, and literature reports of hydrogen peroxide sensitivity in other *Gyrodactylus* species provided additional support for the expansion of the label claim by supporting the likelihood that similar effectiveness will be observed in untested *Gyrodactylus* species. A pdf copy of CVM's notification letter will soon be available on the AADAP website.

PENNOX® 343 (oxytetracycline-HCL)

Potential efficacy studies for immersion therapy— Looking toward the horizon, it's nice to see that we are wrapping up a number of the studies we long ago committed to do in support of drug approvals. However, as we wrap up some commitments, we take on others. As such, we've been having discussions with colleagues around the country about the possibility of conducting studies to evaluate the effectiveness of oxytetracycline hydrochloride to control infestations of susceptible pathogens in a variety of freshwater finfish. Several colleagues have stepped up and have indicated a willingness to help us conduct studies necessary to fulfill the effectiveness data requirements for at least one disease/pathogen (columnaris/*Flavobacterium columnare*). Well, first things first: we developed a research study protocol titled "The Efficacy of PENNOX® 343 (oxytetracycline hydrochloride) Administered as a Static Bath to Control Mortality of Freshwater-Reared Finfish" and submitted it to CVM on July 11, 2012. We hope that by this time next year, we've got a couple of successful effectiveness studies under our belt.

Text provided by Jim Bowker (jim_bowker@fws.gov), Research Program Manager; USFWS AADAP; Bozeman, Montana USA.

FINS & TAILS, BITS & BOBBERS

Level of Anesthesia – Clarification Needed

CVM recently contacted AADAP requesting clarification on how we (and hence y'all) determine if an anesthetic is used to sedate fish to the (1) handleable level or (2) anesthetized level. CVM is reviewing the 2010 Annual INAD Reports for AQUI-S®20E and Benzoak® and has noticed that these terms are being used somewhat interchangeably. In order for us to not only better satisfy data reporting requirements, but also to enhance the future utility of INAD data, we would like to remind our participants of the definitions of each term.

Sedation to Handleable—Use of an anesthetic to sedate fish to a handleable condition typically involves "light sedation" and a relatively short treatment duration (1-5 minutes). A fish is considered handleable when it begins to lose equilibrium, and when it has lost reactivity to most external stimuli with the exception of strong pressure. This condition generally occurs after a fish stops avoiding obstacles in its path and before it completely loses equilibrium. As a general rule, a fish will be considered handleable when it can be held underwater for several seconds without great difficulty. This is similar to Stage 2 anesthesia as described by Sumerfelt and Smith, 1990 (AQUI-S E #11-741 Protocol, pages 8 & 9). Examples of sedation to handleable include treatment to facilitate fin-clipping, collection of length/weight data, and handling for spawning.

Sedation to Anesthetized—Use of an anesthetic to sedate fish to an anesthetized condition typically involves "deep sedation" and a relatively long treatment duration (5-10 minutes). A fish is considered anesthetized when it loses all reflex activity. This

condition generally occurs after a fish has completely lost equilibrium. As a general rule, a fish will be considered anesthetized when it can be easily held out of water, and when lifting the operculum and touching the gill lamellae does not elicit a reflexive "cough" within 5 seconds. This is similar to Stages 4-5 anesthesia as described by Sumerfelt and Smith, 1990 (AQUI-S E #11 -741 Protocol, pages 8 & 9). Examples of sedation to anesthetized include treatment to facilitate implantation of radio transmitters, or other surgical procedures that would require deep sedation and a relatively extended treatment duration.

Please review any anesthetic studies that you may have already entered into the 2012 IPMS on-line database to make sure the correct level of anesthesia has been reported. If you have any questions concerning how you should classify your anesthetic use, please either call or email Bonnie Johnson at 406-994-9905 or bonnie_johnson@fws.gov.

2012 INAD Program Management System (IPMS) Enrollees:

Well……it has already been 6 months since the launch of the IPMS - On-line Data Reporting database, and I wanted to give a brief update on where we are at. First, we have over 360 study monitor and investigator accounts currently set up in the system and over 450 studies in various stages of completion. We have definitely been "putting the system to the test," and although overall it has been working fairly smoothly, we are most certainly aware that a number of recurring "bugs" still exist. Please rest assured that we are working diligently to eliminate these issues. In the interim, provided below are several "helpful hints" that should be beneficial both now and in the future:

(1) It has come to my attention that some investigators are not aware that the information that is carried over from the Study Request (Stage 1) should be edited in the Study Results Form (Stage 4). Please be sure to review and edit (if necessary) all data that is carried over from the Study Request and ensure the data fields reflect the actual information (i.e., Total Number of Treated Fish; Drug Dose; Disease Treated/Treatment Objective, etc.).

(2) Conflicts in number of treated fish and/or treatment dates have been a recurring problem within the IPMS. It is important to note that such information is requested on multiple forms (locations) within the IPMS, and all entries must "match" or errors will be noted. Please be sure that all treatment information is consistent throughout all study forms.

(3) Please note studies should be submitted to the AADAP Office within 30 days of completion. If your study (or one for which you are the Study Monitor) appears to be stuck at a given stage, please contact the appropriate person so they can log into their account and move the study to the next stage. Because we have experienced some issues with automatic email notifications of study events, it could be that folks are simply not always aware that their attention/action is required.

(4) The User Manual is located on the menu bar located at the far right. Please review this manual because it should provide you with helpful information on how to navigate the IPMS on-line database.

Sponsorship of sGnRHa (OvaRH®) INAD submitted to CVM

The AADAP Office has recently submitted a request to FDA for INAD authorization for the use of sGnRHa (OvaRH®) as an injectable treatment to induce final gamete maturation (ovulation and spermiation) in freshwater and marine finfish. sGnRHa (OvaRH®) is an injectable product that has been developed by Western Chemical, Inc., specifically to induce gamete maturation in a broad variety of fish species. AADAP is requesting an authorization to slaughter (i.e., harvest) treated animals and the establishment of a withdrawal time under this new INAD. We anticipate this INAD should be in place before next year's spawning season. We will keep you updated on the progress of this new INAD!

Text provided by Bonnie Johnson (bonnie_johnson@fws.gov), National INAD Program Administer, USFWS AADAP; Bozeman, Montana USA

FEATURE ARTICLE

Introducing the U.S. Food and Drug Administration Center for Veterinary Medicine Office of Research

by
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The Office of Research (OR) supports the Center for Veterinary Medicine's (CVM) mission to regulate veterinary drugs, feeds and devices given to animals. The Office is located in Laurel, Maryland, at an FDA research facility consisting of two main research buildings, laboratory and farm animal outbuildings, a small dairy, approximately 38 acres of pasture, and an aquaculture building. The Office employs animal scientists, biologists, chemists, pharmacologists, immunologists, microbiologists, epidemiologists and veterinarians. The Division of Animal and Food Microbiology studies how antimicrobial use in animals

impacts drug efficacy against microbial pathogens, changes in the environmental microbial ecology, and the development of antimicrobial resistance in pathogenic and commensal microorganisms. The Division of Residue Chemistry (DRC) develops and validates analytical detection methods for compounds which pose a potential health risk if found in animal tissue or feed. The Division of Applied Veterinary Research (DAVR) conducts research using animals and animal-derived systems to study the fate of veterinary drugs, feed additives, or toxicants in animals. In addition, DAVR develops and validates tests to confirm the identity of animal food products, and to detect prohibitive animal material and compounds in animal feeds.

Aquaculture research at OR is designed to assist FDA in ensuring that fish derived from aquaculture production environments (domestic or international) is safe for human consumption. These activities fall into two categories: (1) development of chemical detection methods (led by DRC), and (2) animal and microbiology studies (led by the DAVR Aquaculture Team). Research priorities explore drug residue persistence and distribution, metabolism, efficacy, and environmental effects of drugs and other chemicals used in aquaculture. Capabilities include:

- Developing disease models to test drug effects,
- Pharmacokinetics studies (how fish distribute, metabolize and eliminate drugs),
- Depletion studies of prohibited chemicals (e.g., feed contaminants),
- Detection and identification of aquatic pathogens,
- Development of methods to test antibiotic susceptibility of aquatic bacteria, and
- Generating tissues incurred with drug residues to validate detection methods.

Highlights of current aquaculture research at OR are:

1. Development of antimicrobial susceptibility testing methods.

The DAVR Aquaculture Team has led in the development of standard methods to test the antimicrobial susceptibility of bacteria isolated from aquatic animals. In response to CVM's recent fish drug approvals to limit losses from the gliding aquatic bacteria *Flavobacterium columnare* and *F. psychrophilum*, the DAVR Aquaculture Team has developed reliable susceptibility testing procedures for these bacteria and completed a multi-laboratory testing trial to establish quality control limits to standardize these tests. These methods are being added to a testing guideline published by the Clinical Laboratory Standards Institute (CLSI).

2. Development of chemical detection methods.

DRC chemists are currently developing a single analytical method that detects an array of drugs in shrimp, a single analytical method that detects multiple hormone treatments across different food fish species, and a chemical assay to detect erythromycin in medicated fish feeds. DRC also leads an interagency working group that coordinates the development of detection methods for FDA's regulatory surveillance of seafood.

3. *In vitro* screening of drug effects on a fish monogenean parasite.

DAVR biologists recently developed laboratory techniques to demonstrate and evaluate drug effects on a monogenean parasite of largemouth bass to help demonstrate how to test drug effects on fish parasites. The method uses video microscopy to visually document the altered behavior of the parasite to seven different antiparasitic drugs. Only one antiparasitic drug is approved for use in fish; therefore, a screening method to identify drug effects is beneficial to potential sponsors seeking to identify possible candidate drugs.

4. Pharmacokinetic studies.

OR scientists have compared how fish metabolize the antiparasitic drugs ivermectin and albendazole across a range of farmed fish species. This cross-species comparison helps determine whether the marker residue identified in mammals to monitor drug use is valid for detecting those drugs in various fish species. The marker residue is the active form of the drug (original compound or metabolite) that lingers longest in the animal's body. It provides regulators the best chance to find potential illegal drug residues in food animal products.

In addition, the DAVR Aquaculture Team has constructed the Phish-Pharm database. This is a Microsoft Access-based searchable compilation of more than 520 articles from the peer-reviewed literature dealing with drug residues and pharmacokinetic parameters in over 124 species (95 genera) of fish. A more basic version is also available as a web-based searchable page. Phish-Pharm can facilitate the study of aquatic species' drug metabolism and can rapidly compare data between studies with different experimental conditions, such as water temperatures and salinity. The database is regularly updated and is readily available to use and download at: *www.fda.gov/AnimalVeterinary/ScienceResearch/ToolsResources/Phish-Pharm/default.htm*

5. Feed contaminant studies.

Office of Research scientists have also responded to emergency situations such as the melamine/cyanuric acid pet food contamination and recall event of 2007, which included commercial agriculture and aquaculture

feeds. Fish were used as sentinels in studies conducted by OR to investigate how animals eliminate and sequester melamine and cyanuric acid from muscle (edible portions), kidneys (the target organ of toxicity) and serum.

In conclusion, the expertise at OR provides a valuable resource to answer questions that arise during the review of new animal drug application and to develop technologies that improve the drug review process. Our aquaculture program actively collaborates with the CVM Office of New Animal Drug Evaluation and Office of Surveillance and Compliance, scientists at other FDA centers and outside agencies such as the USDA, the University of Maryland, The Johns Hopkins University, and the Maryland Department of Natural Resources.

USGS's UMESC CORNER

Sedatives

The U.S. Geological Survey's (USGS) Upper Midwest Environmental Sciences Center (UMESC) completed a study to determine the exposure parameters (concentration and duration) that will maximize eugenol residues in the fillet tissue from rainbow trout exposed to AQUI-S®20E (active ingredient, eugenol) and determine the sample times that will adequately characterize the depletion of eugenol residues from the fillet tissue of exposed fish. Fish were exposed to 50 mg/L AQUI-S®20E for various times through 1,440 min, to 100 and 250 mg/L for various times through 240 min, and to 500 and 1,000 mg/L for various times through 90 min. Fillet tissue concentrations were maximized after exposure to 100 mg/L AQUI-S®20E for 30, 60, 120, and 240 min. Eugenol concentrations were 50, 58, 54, and 62 µg/g, respectively. All other exposure concentrations and durations resulted in eugenol concentrations <39 µg/g. To assess eugenol depletion, rainbow trout were exposed to 100 mg/L AQUI-S®20E for 60 min when they were transferred to fresh, flowing water and thereafter sacrificed at various times through 1,440 min. Eugenol concentrations in fillet tissues were 19.7, 4.1, and 0.4 µg/g after 30, 120, and 360 min of depletion, respectively. The final report is scheduled to be submitted to the FDA Center for Veterinary Medicine in August 2012.

UMESC has made an intense effort to gear up to conduct a total residue depletion study with ^{14}C eugenol where the depletion, distribution, and identification of eugenol residues in the fillet tissue will be characterized. Synthesis of the radioactive eugenol is nearly complete, and delivery is expected mid to late August. After receipt of the chemical, UMESC will conduct a pilot study to ensure analysts are adequately trained in all technical procedures for exposing fish and processing fillet tissue from exposed fish. Thereafter, the definitive study will be conducted. The pilot study is scheduled to begin in late August 2012, and the definitive study is scheduled to begin in late September 2012.

AQUAFLOR® (50% florfenicol)

UMESC conducted a study that fulfilled the following objectives: (1) determine the depletion rate of the florfenicol amine (FFA) residues from the fillet tissue of rainbow trout treated with florfenicol (FFC)-medicated feed in a recirculating aquaculture system, (2) determine the FFC concentrations in the water of the recirculating aquaculture system during and after treating rainbow trout with FFC-medicated feed, (3) determine FFA residue concentrations in the fillet tissue of nontreated rainbow trout sharing a recirculating aquaculture system with rainbow trout treated with FFC-medicated feed, and (4) determine the depletion rate of FFA from the fillet tissue of rainbow trout treated with FFC-medicated feed in a flow-through aquaculture system. Analyses of fillet tissue from treated fish are scheduled to be completed in August 2012. Construction of a final report is underway. A draft of the final report is scheduled to be completed in October 2012.

Text provided by Jeff Meinertz (jmeinertz@usgs.gov), Research Physiologist; USGS UMESC; La Crosse, Wisconsin USA.

USDA's ARS CORNER

Aquaculture America 2012 and 2013

At Aquaculture America 2012 (Feb 28-Mar 2, Las Vegas, Nevada USA), the Aquaculture Drug Research and Drug Approval Status special session had 12 presentations with great attendance for each presentation. The session was organized and moderated by Jim Bowker and Dr. Dave Straus, and this was the 10th year we have held this session focused on research in aquaculture therapeutants.

Currently, we are soliciting speakers for Aquaculture America 2013 (Feb 21-25, Nashville, Tennessee USA). If you are interested in presenting, contact Jim Bowker (jim_bowker@fws.gov) or Dr. Dave Straus (dave.straus@ars.usda.gov).

Copper Sulfate ($CuSO_4$)

Two Final Study Reports covering the pivotal effectiveness dose-confirmation study of $CuSO_4$ on fungus of channel catfish eggs have been sent to the sponsor and should be submitted to FDA by the time you read this. This should complete all technical sections except for Environmental Safety.

Also, we recently completed a study where we held catfish in water that was treated with $CuSO_4$ for 24 h and then looked at fish resistance to columnaris disease after 0 h, 24 h, and 9 d. We challenged the fish in our low-flow aquarium system and found fish were significantly resistant at 24 h and 9 d, but not a 0 h. This information is useful for transferring fingerlings to ponds

or grading fish.

Peracetic Acid (PAA)

Our work with peracetic acid (PAA) continues; we have completed studies on the effectiveness of PAA to control fungus on eggs and to determine the LC50 and resulting histopathology of catfish fry exposed to PAA. We found that the optimum flow-through treatment rate for PAA was 5 mg/L to control egg fungus, but 2.5 mg/L gives a greater margin of safety to hatching fry. In the toxicity study, we found that yolk-sac fry were more tolerant of PAA than swim-up fry by 1.4-fold (24/48 h LC50 values were 2.6 vs. 1.8 mg/L PAA). An advantage to using PAA includes very low environmental impact considerations, as it degrades to harmless residues rapidly.

Text provided by Dr. Dave Straus (dave.straus@ars.usda.gov), Research Toxicologist; U.S. Department of Agriculture, Agricultural Research Service; Harry K. Dupree – Stuttgart National Aquaculture Research Center; Stuttgart, Arkansas USA.

AFS's WGADCB CORNER

The American Fisheries Society's (AFS) Fish Culture Section (FCS) Working Group on Aquaculture Drugs, Chemicals, and Biologics (WGADCB) met February 29, 2012, during Aquaculture America 2012 (Las Vegas, Nevada USA) and July 31, 2012, during the 18th Annual USFWS Aquaculture Drug Approval Coordination Workshop (La Crosse, Wisconsin USA). Meeting highlights included the following:

● Updates to the AFS publication, *Guide to Drug, Vaccine, and Pesticide use in Aquaculture*, have been posted online and include updates to the approved uses of AQUAFLOR® (50% florfenicol) and two new vaccines (*https://sites.google.com/site/fishculturesection/home*). The next revision cycle is planned for the end of 2012; however, sponsors and other interested parties are encouraged to submit updates to the WGADCB as they become available. At least one sponsor has expressed interest in being able to distribute the *Guide* to their customers to encourage proper use of the sponsor's products. The WGADCB supports this idea, and other interested sponsors are encouraged to contact the WGADCB.

● Leading representatives of AFS, the U.S. Food and Drug Administration (FDA) Center for Veterinary Medicine (CVM), and the fish drug research and development community met this spring in Rockville, Maryland USA, to discuss the AFS policy statement on the need for immediate-release sedatives and other issues related to the approval and use of aquaculture drugs in the United States. The meeting helped facilitate the amended authorization allowing the immediate-release use of AQUI-S®20E for field use under USFWS INAD-11-741. Also, an article titled, e.g., *Aquaculture Drug FAQ with FDA,* is planned for the AFS publication *Fisheries*.

● A draft of the FDA document, *FDA Guidance Document 61: Guidance For Industry, FDA Approval of New Animal Drugs for Minor Uses and Minor Species*, is being reviewed within FDA before being released for public comment. This document might not be released for public comment until sometime in 2013.

● As a result of a "Listening Session" held with FDA CVM in summer 2011, several *ad-hoc* groups are continuing to work on special topics (e.g., strategies to support pathogen grouping). Currently, there are no plans to hold a second session until at least some of the issues identified in the first session have been adequately addressed.

● One constraint to end-users purchasing approved aquaculture drug products is that less expensive—but unapproved—products are sometimes available. Some public agencies are required to purchase goods and services from the lowest bidding contractor, and such a requirement presents a conflict every time drug purchases are made. A letter from FDA to public agencies that explains the need to purchase only approved products could help address the concerns of purchasing agents—particularly if such a letter could be made available online so that it could be printed and included with every purchase request.

● There is interest in having access to an online, wish-list of studies that could be conducted to support aquaculture drug approvals (e.g., label expansions). It might be possible to maintain such a list on a public agency website (e.g., USGS Upper Midwest Environmental Science Center website or USFWS Aquatic Animal Drug Approval Partnership program website). Periodic reminders could be distributed to potential collaborators by the WGADCB.

Meeting notes provided by Dr. Jesse Trushenski (saluski@siu.edu), Assistant Professor; Southern Illinois University at Carbondale; Fisheries and Illinois Aquaculture Center; Carbondale, Illinois USA and Jim Bowker (jim_bowker@fws.gov), Research Program Manager; USFWS AADAP; Bozeman, Montana USA. Meeting notes edited for this newsletter by Dan Carty, USFWS AADAP.

FDA's CVM NOTES

Personnel Updates

The U.S. Food and Drug Administration (FDA) Center for Veterinary Medicine's (CVM) Dr. Joan Gotthardt and Chuck Eirkson are familiar to many of this newsletter's readers, and so we wanted to let you know of their retirements. While Joan was most recently a member of the Office of Minor Use and Minor Species, previously

she had served as the Director of the Division of Therapeutic Drugs for Food Animals in the Office of New Animal Drug Evaluation (ONADE), and before that, as the Leader of the Aquaculture Drugs Team. Chuck was the Leader of the Environmental Safety Team in ONADE. We will miss them!

Minor use and minor species questions can be directed to Dr. Meg Oeller (Director), Dr. Stuart Jeffrey (questions related to designation), and Dr. Dorothy Bailey (questions related to indexing). Currently, Dr. Eric Silberhorn is the Acting Leader of the Environmental Safety Team, and Dr. Veronica Taylor is the Acting Director of the Division of Scientific Support Staff in ONADE. The Division of Scientific Support Staff includes the Biometrics Team and the Environmental Safety Team. Contact information for these folks, and other CVM staff, can be accessed through the following webpage:

http://www.fda.gov/aboutfda/centersoffices/officeoffoods/cvm/default.htm.

Text provided by Dr. Jennifer Matysczak (Jennifer.Matysczak@fda.hhs.gov), Leader; Aquaculture Drugs Team; Office of New Animal Drug Evaluation; Center for Veterinary Medicine, Food and Drug Administration; Rockville, Maryland USA

RELEVANT LITERATURE

Listed below are journal citations with particular relevance to the broad topic of drugs and aquaculture species. With some exceptions, this list includes citations not previously included in our newsletter. Our complete Relevant Literature list, which dates back to 2009, can be viewed or downloaded by clicking here.

Inclusion of a citation in our newsletter does not imply (1) acceptance by the U.S. Food and Drug Administration's Center for Veterinary Medicine of a drug's safety or effectiveness, (2) endorsement of a drug or product by the U.S. Fish & Wildlife Service, (3) recommendation of the technique to any particular situation, or (4) concurrence with a treatment procedure/ drug.

If you know of literature that might be of interest to our readers, please send the citation to Dan Carty. *dan_carty@fws.gov*

Antibiotic and Bacterial

Can, E, et al. 2012. Ozone disinfection of eggs of gilthead seabream *Sparus aurata*, sea bass *Dicentrarchus labrax*, red porgy, and common dentex *Dentex dentex*. *Journal of Aquatic Animal Health* **24(2):129-133**.

Carraschi, SP, et al. 2012. Histopathological biomarkers in pacu (*Piaractus mesopotamicus*) infected with *Aeromonas hydrophila* and treated with antibiotics. *Ecotoxicology and Environmental Safety* **83:115-120**.

Castillo, D, et al. 2012. Diversity of *Flavobacterium psychrophilum* and the potential use of its phages for protection against bacterial cold water disease in salmonids. *Journal of Fish Diseases* **35(3):193-201**.

Chang, Z-Q, et al. 2012. The effect of temperataure and salinity on the elimination of enrofloxacin in the Manila clam *Ruditapes philippinarum*. *Journal of Aquatic Animal Health 24(1):17-21*.

Chen, M-H, et al. 2012. *Lactococcus lactis* subsp. *lactis* infection in Bester sturgeon, a cultured hybrid of *Huso huso* × *Acipenser ruthenus*, in Taiwan. *Research in Veterinary Science* **93(2):581-588**.

Chen, YF, et al. 2012. Isolation and characterization of *Aeromonas schubertii* from diseased snakehead, *Channa maculata* (Lacepède). *Journal of Fish Diseases* **35(6):421-430**.

Deng, B, et al. 2012. Pharmacokinetics and residues of tetracycline in crucian carp muscle using capillary electrophoresis on-line coupled with electrochemiluminescence detection. *Food Chemistry* **134(4):2350-2354**.

Farmer, BD, et al. 2012. Effectiveness of copper sulfate and potassium permanganate on channel catfish infected with *Flavobacterium columnare*. *North American Journal of Aquaculture* **74(3):320-329**.

Figueiredo, HCP, et al. 2012. *Weissella* sp. outbreaks in commercial rainbow trout (*Oncorhynchus mykiss*) farms in Brazil. *Veterinary Microbiology* **156(3-4):359-366**.

Geng, Y, et al. 2012. *Streptococcus agalactiae*, an emerging pathogen for cultured ya-fish, *Schizothorax prenanti*, in China. *Transboundary and Emerging Diseases* **59(4):369-375**.

Henríquez-Núñez, H, et al. 2012. Antimicrobial susceptibility and plasmid profiles of *Flavobacterium psychrophilum* strains isolated in Chile. *Aquaculture* **354-355:38-44**.

Hurtado de Mendoza, J, et al. 2012. Validation of antibiotics in catfish by on-line solid phase extraction coupled to liquid chromatography tandem mass spectrometry. *Food Chemistry* **134(2):1149-1155**.

Imanpoor, MR, et al. 2011. Effects of sublethal concentration of chloramine-T on growth, survival, haematocrit and some blood biochemical parameters in common carp fry (*Cyprinus carpio*). *Aquaculture, Aquarium, Conservation & Legislation* **4(3):280-291**.

Lee, D-K, et al. 2012. Antibiograms and the estimation of epidemiological cut off values for *Vibrio*

ichthyoenteri isolated from larval flounder, *Paralichthys olivaceus*. *Aquaculture* **342-343:31-35**.

Madhuri, S, et al. 2012. Antimicrobial activity of some medicinal plants against fish pathogens. *International Research Journal of Pharmacy* **3(4):28-30**. (**bacteria and fungi**).

Miller, RA, et al. 2012. Oxytetracyline pharmacokinetics in rainbow trout during and after an orally administered medicated feed regimen. *Journal of Aquatic Animal Health* **24(2):121-128**.

Munasinghe, N, et al. 2012. Farm level and geographic predictors of antibiotic use is Sri Lankan shrimp farms. *Journal of Aquatic Animal Health* **24(1):22-29**.

Nair, AV, et al. 2012. Diversity and characterization of antagonistic bacteria from tropical estuarine habitats of Cochin, India for fish health management. *World Journal of Microbiology & Biotechnology* **28(7):2581-2592**.

Oplinger, RW, and Wagner, EJ. 2012. Effects of media ingredient substitution and comparison of growth of *Flavobacterium psychrophilum* among four media. *Journal of Aquatic Animal Health* **24(1):49-57**.

Ostrand, SL, et al. 2012. Inhibitory effects of rosemary oil on the *in vitro* growth of six common finfish pathogens. *North American Journal of Aquaculture* **74(2):230-234**.

Smith, EM, et al. 2012. *In vitro* inhibition of cytochrome P450-mediated reactions by gemfibrozil, erythromycin, ciprofloxacin and fluoxetine in fish liver microsomes. *Aquatic Toxicology* **109:259-266**.

Touraki, M, et al. 2012. Treatment of vibriosis in European sea bass larvae, *Dicentrarchus labrax* L., with oxolinic acid administered by bath or through medicated nauplii of *Artemia franciscana* (Kellogg): efficacy and residual kinetics. *Journal of Fish Diseases* **35(7):513-522**.

Vendrell, D, et al. 2012. Accumulation and depletion kinetics of erythromycin in rainbow trout (*Oncorhynchus mykiss*). *Preventive Veterinary Medicine* **105(1-2):160-163**.

Wang, H, et al. 2012. Maternal transfer and protective role of antibodies in zebrafish *Danio rerio*. *Molecular Immunology* **51(3-4):332-336**.

Zahran, E, et al. 2012. The effect of adjuvant and microbial challenge on the expression of antimicrobial polypeptides in channel catfish (*Ictalurus punctatus*). *Fish & Shellfish Immunology* **33(2):168-173**.

Parasite and Fungus Control

Akoll, P, et al. Risk assessment of parasitic helminthes on cultured Nile tilapia (*Oreochromis niloticus*, L.). *Aquaculture* **356-357:123-127**.

Barnes, JM, et al. 2012. Initial investigations of hops as a salmonid egg fungicide. *North American Journal of Aquaculture* **74(3):310-333**.

Bowker, JD, et al. 2012. Efficacy of 35% PEROX-AID (hydrogen peroxide) to reduce an infestation of *Gyrodactylus salmonis* in rainbow trout. *North American Journal of Aquaculture* **74(2):154-159**.

Bowker, JD, et al. 2012. Efficacy of SLICE premix (0.2% emamectin benzoate) for reducing infestations of *Salmincola* spp. in freshwater-reared rainbow trout. *North American Journal of Aquaculture* **74(3):428-437**.

Budiño, B, et al. 2012. Differences in the *in vitro* susceptibility to resveratrol and other chemical compounds among several *Philasterides dicentrarchi* isolates from turbot. *Parasitology Research* **110 (4):1573-1578**.

Caruana, S, et al. 2012. The efficacy of selected plant extracts and bioflavonoids in controlling infections of *Saprolegnia australis* (Saprolegniales; Oomycetes). *Aquaculture* **358-359:146-154**

Gunn, C, et al. 2012. Pilot field trial to evaluate SLICE (0.2% emamectin benzoate)-medicated feed to reduce a natural infestation of *Salmincola californiensis* in rainbow trout. *North American Journal of Aquaculture* **74(3):424-427**.

Heumann, J, et al. 2012. Molecular cloning and characterisation of a novel P-glycoprotein in the salmon louse *Lepeophtheirus salmonis*. *Comparative Biochemistry and Physiology – Part C: Toxicology & Pharmacology* **155(2):198-205**.

Horsberg, TE. 2012. Avermectin use in aquaculture. *Current Pharmaceutical Biotechnology* **13(6):1095-1102**. **(review article)**

Igboeli, OO, et al. 2012. Role of P-glycoprotein in emamectin benzoate (SLICE®) resistance in sea lice, *Lepeophtheirus salmonis*. *Aquaculture* **344-349:40-47**.

Jimenez, DF, et al. 2012. Confidence in assessing the effectiveness of bath treatments for the control of sea lice on Norwegian salmon farms. *Aquaculture* **344-349:58-65**.

Kawano, F, and Hirazawa, N. 2012. Antiparasitic effect of in-feed inhibitors of folic acid synthesis and dihydrofolate reductase against ciliate *Cryptocaryon irritans* infection in the red sea bream *Pagrus major* and against ciliate *Ichthyophthirius multifiliis* infection in black pop-eyed goldfish *Carassius auratus*. *Aquaculture* **330-333:1-7**.

Kawano, F, et al. 2012. Antiparasitic effects of dietary Romet 30 (SDMX-OMP) against ciliate *Cryptocaryon irritans* infection in the red sea bream *Pagrus major* and tiger puffer *Takifugu rubripes*. *Aquaculture* **344-349:35-39**.

Larrat, S, et al. 2012. Safety and efficacy of emamectin benzoate to treat *Anguillicoloides crassus* (Kuwahara, Niimi & Itagaki) infections in American eels, *Anguilla rostrata* (Lesueur). *Journal of Fish Diseases* **35(6):467-470**.

Pahor-Filho, E, et al. 2012. Parasitology of juvenile mullet (*Mugil liza*) and effect of formaldehyde on parasites and host. *Aquaculture* **354-355:111-116**.

Picón-Camacho, SM, et al. 2012. Effects of long duration, low dose bronopol exposure on the control of *Ichthyophthirius multifiliis* (Ciliophora), parasitising rainbow trout (*Oncorhynchus mykiss* Walbaum). *Veterinary Parasitology* **186(3-4):237-244**.

Porter, J., et al. 2012. Development of an evidence biochip array kit for the multiplex screening of more than 20 anthelmintic drugs. *Analytical & Bioanalytical Chemistry* **403(10):3051-3056**.

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UPCOMING MEETINGS

USTFA 2012 (September 6-8, Denver, Colorado USA): The United States Trout Farmers Association (USTFA) 2012 Fall Conference will be held September 6-8 in Denver, Colorado USA (http://www.ustfa.org/industry/events/events.html).

The conference will include a half-day workshop on *Aquaculture Business Management and Marketing*, several technical sessions, a USTFA business meeting, a trade show, socials and a raffle, and a field tour of Rocky Mountain National Park and Liley Fisheries (an aquatic habitat and fisheries consulting firm).

Aqua SUR 2012 (October 10-13, Puerto Montt, Chile): AQUA SUR 2012—the most important international aquaculture fair in the southern hemisphere—will be held October 10-13 in Puerto Montt, the capital of Chilean salmon (http://www.aqua-sur.cl/2012/feria_en.php).

AQUA SUR will bring together exhibitors from the five continents and over 40 countries to show their products and acquire and exchange knowledge. It is an excellent opportunity to know and roam around the aquaculture industry in a single place and to learn about the latest developments in products, services, and technologies that are taking place in Chile and globally.

Offshore Mariculture 2012 (October 17-19, Izmir, Turkey): The Offshore Mariculture 2012 conference will be held October 17-19 in Izmir, Turkey (http://www.offshoremariculture.com).

Offshore Mariculture 2012 is an international two-day conference focusing on the offshore fish farming business. The conference will explore the progress and prospects for offshore aquaculture in European and international waters. The two day technical conference will be followed by a third day dedicated to a visit to an offshore fish farm.

Northwest Fish Culture Conference 2012 (December 11-13, Portland, Oregon USA): The U.S. Fish and Wildlife Service and the Oregon Chapter of the American Fisheries Society will be hosting the 63rd Annual Northwest Fish Culture Conference on December 11-13. (http://www.fws.gov/columbiariver/nwfcc2012.html). If you are interested in making an oral presentation or presenting a poster, please contact Nathan Wiese at nathan_wiese@fws.gov or Jim Bowker at jim_bowker@fws.gov.

Northeast Aquaculture Conference 2012 (December 12-15, Groton, Connecticut USA: Please plan to attend this special joint meeting of the Northeast Aquaculture Conference & Exposition (NACE), Milford Aquaculture Seminar (MAS), and International Conference

on Shellfish Restoration (ICSR) on December 12-15 in Groton, Connecticut USA (*http://www.northeastaquaculture.org*). This biennial conference and trade show brings shellfish, finfish and algae producers together with industry vendors, regulators and NGOs to discuss the latest research and issues affecting the industry.

Texas Aquaculture Association Conference and Trade Show 2013 (January 23-25, 2013, Bay City, Texas): Visit the conference website at *http://www.texasaquaculture.org/*.

AQUACULTURE 2013 (February 21-25, Nashville, Tennessee USA): The World Aquaculture Society will hold its Triennial meeting in the exciting city of Nashville, Tennessee USA, on February 21-25, 2013 (*https://www.was.org/WasMeetings/meetings/Default.aspx?code=AQ2013*).

The Triennial is the largest aquaculture conference and tradeshow in the world, with nearly 4,000 attendees from over 90 countries. The Triennial combines the annual meetings of the Fish Culture Section of the American Fisheries Society, the World Aquaculture Society, and the National Shellfisheries Assoc.

In addition to these meetings, look what else is happening at AQUACULTURE 2013:

● AQUACULTURE AMERICA - Annual Meeting of the U.S. Chapter of World Aquaculture Society, the National Aquaculture Assoc., and the U.S. Aquaculture Suppliers Assoc.
● Annual Meeting of the American Tilapia Assoc.
● Annual Meeting of the Striped Bass Growers Assoc.
● Annual Meeting of the U.S. Trout Farmers Assoc.
● Special sessions organized by the Aquacultural Engineering Society and International Association of Aquaculture Economics and Management
● Many other meetings of work groups, government agencies, and related aquaculture activities