The Open Access Israeli Journal of Aquaculture – Bamidgeh

As from **January 2010** The Israeli Journal of Aquaculture - Bamidgeh (IJA) will be published exclusively as **an on-line Open Access (OA)** quarterly accessible by all AquacultureHub (http://www.aquaculturehub.org) members and registered individuals and institutions. Please visit our website (http://siamb.org.il) for free registration form, further information and instructions.

This transformation from a subscription printed version to an on-line OA journal, aims at supporting the concept that scientific peer-reviewed publications should be made available to all, including those with limited resources. The OA IJA does not enforce author or subscription fees and will endeavor to obtain alternative sources of income to support this policy for as long as possible.

Editor-in-Chief
Dan Mires

Editorial Board

Sheenan Harpaz	**Agricultural Research Organization Beit Dagan, Israel**
Zvi Yaron	**Dept. of Zoology Tel Aviv University Tel Aviv, Israel**
Angelo Colorni	**National Center for Mariculture, IOLR Eilat, Israel**
Rina Chakrabarti	**Aqua Research Lab Dept. of Zoology University of Delhi**
Ingrid Lupatsch	**Swansea University Singleton Park, Swansea, UK**
Jaap van Rijn	**The Hebrew University Faculty of Agriculture Israel**
Spencer Malecha	**Dept. of Human Nutrition, Food and Animal Sciences University of Hawaii**
Daniel Golani	**The Hebrew University of Jerusalem Jerusalem, Israel**
Emilio Tibaldi	**Udine University Udine, Italy**

Copy Editor
Ellen Rosenberg

Published under auspices of
The Society of Israeli Aquaculture and Marine Biotechnology (SIAMB),
University of Hawaii at Manoa Library
and
University of Hawaii Aquaculture Program in association with
AquacultureHub
http://www.aquaculturehub.org

ISSN 0792 - 156X

© Israeli Journal of Aquaculture - BAMIGDEH.

PUBLISHER:
Israeli Journal of Aquaculture - BAMIGDEH -
Kibbutz Ein Hamifratz, Mobile Post 25210,
ISRAEL
Phone: + 972 52 3965809
http://siamb.org.il

Comparative Gene and QTL Mapping in Aquaculture Species

Gary H. Thorgaard[1]*, Krista M. Nichols[2] and Ruth B. Phillips[3]

[1] *School of Biological Sciences and Center for Reproductive Biology,*
Washington State University, Pullman, WA 99164-4236 USA

[2] *Departments of Biological Sciences & Forestry and Natural Resources, Purdue University,*
West Lafayette, IN 47907 USA

[3] *School of Biological Sciences and Center for Reproductive Biology,*
14204 NE Salmon Creek Ave., Washington State University, Vancouver, WA 98686-9600 USA

Key words: QTL mapping, comparative mapping, aquaculture, genomics, sequencing

Extended Abstract

Mapping genes and quantitative trait loci (QTLs) is of fundamental and applied interest in aquaculture species. Such studies can provide essential information about rates and patterns of evolutionary change and may provide tools which can be used in marker-assisted selection (MAS; e.g., Groenen et al., 2000). Mapping typically involves producing divergent crosses and analyzing segregation patterns in the offspring of hybrids between the divergent crosses. The more divergent the cross is, the greater the potential for finding useful marker and trait differences that can be mapped. The basic goal of mapping is to understand the position of loci on chromosomes and how the positions change during evolution. The applied goal of mapping is to understand the position of loci associated with phenotypic traits which could be important in breeding programs. Correspondingly, QTL mapping seeks to identify the number and location of loci associated with phenotypic traits (e.g., Mackay, 2001; Mauricio, 2001; Burt and Hocking, 2002; Erickson et al., 2004) while applied QTL studies seek to identify candidate genes or markers for use in MAS.

More detailed DNA sequence information is available in model research species (e.g., fugu, zebrafish, medaka, stickleback) than in aquaculture species (e.g., rainbow trout, Atlantic salmon, channel catfish, tilapia, Japanese flounder, oyster and shrimp; Clark, 2003). Genetic maps are relatively well-developed for several research model species (e.g., Woods et al., 2000; Naruse et al., 2004; Kai et al., 2005) and aquaculture species (Kocher et al., 1998; Young et al., 1998; Sakamoto et al., 2000; Waldbieser et al., 2001; Wilson et al., 2002; Coimbra et al., 2003; Nichols et al., 2003; Yu and Guo, 2003; Moen et al., 2004; Danzmann et al., 2005; Rexroad et al., 2005). The model species tend to have better maps for known genes.

QTL information, however, is best developed in the rainbow trout and tilapia. A number of studies have been conducted in rainbow trout using outbred populations (e.g., Sakamoto et al., 1999; Ozaki et al., 2001;

---

* Corresponding author. Tel.: +1-509-335-7438, fax: +1-509-335-3184,
e-mail: thorglab@wsu.edu

Perry et al., 2001, 2005; O'Malley et al., 2002; Martyniuk et al., 2003) and doubled haploid offspring of crosses between clonal lines (e.g., Robison et al., 2001; Nichols et al., 2004; Zimmerman et al., 2004). QTLs for aquaculturally important traits have been studied in crosses in tilapia (e.g., Cnaani et al., 2003, 2004). The genetic architecture of morphological traits is also being studied in related cichlid fishes (e.g., Albertson et al., 2003). QTL studies on model research species include morphological features in sticklebacks (e.g., Peichel et al., 2001; Shapiro et al., 2004) and behavioral and morphological traits in zebrafish (Wright et al., 2006). In some cases, candidate genes are beginning to be associated with specific QTLs. Leder et al. (2006) localized the Clock gene to a QTL region for spawning time and development rate in rainbow trout.

There is considerable interest in being able to extrapolate sequence and map information from model research species to aquaculture species, and within and among evolutionarily important groups. Information on the rate of change in the order of genes on chromosomes across fish species is beginning to become available. The comparison of gene order between the medaka and the zebrafish is the most complete comparison to date (Naruse et al., 2004). Across species, gene order appears to be conserved in large blocks. Within salmonid species, map comparisons among Atlantic salmon, rainbow trout, and arctic char have been made (Danzmann et al., 2005). A recent study expanded the comparison to brown trout (Gharbi et al., 2006). Further linkage and physical mapping of known genes will facilitate comparisons among and within species.

Gene order should be largely conserved within species. Studies on map order within a species, with a few exceptions, should be able to be extrapolated to other populations. If QTLs are conserved across populations within species, it would greatly facilitate the application of QTLs in MAS. The degree to which QTLs are conserved among populations within and across species is beginning to be studied (Paterson et al., 1995; Georges, 1997; Burke et al., 2002; Reid et al., 2005). Grass species have been relatively well-studied for QTL conservation. Examples of conserved QTLs in grasses include those for plant height and maturity (Lin et al., 1995), disease resistance (Chen et al., 2003), and flowering time (Chardon et al., 2004). Vegetative propagation QTLs appear to be conserved across *Eucalyptus* species (Marques et al., 2002) and QTLs were found to be conserved across pepper species (Ben Chaim et al., 2001; Rao et al., 2003). Wood density and cell wall component QTLs show conservation between maritime and loblolly pine (Chagne et al., 2003). A QTL related to blood pressure was found to be conserved between rats and humans (Zimdahl et al., 2002). Recently, Reid et al. (2005) found indications of conservation of body weight QTLs across salmonid fish species.

Highly divergent crosses are often ideal for mapping and identification of QTLs (Andersson, 2001). Examples include the Meishan x Gottingen cross in pigs (Wada et al., 2000) and the white leghorn x jungle fowl cross in chickens (Groenen et al., 2000). Divergent crosses should be easy to produce for many fish species because both domesticated and wild forms from multiple geographic sources are often available. However, these types of crosses have not yet been widely used in fishes.

Divergent crosses between homozygous clonal lines appear to be especially favorable material for mapping and identification of QTLs in fishes. Clonal lines have been successfully produced for a number of fish species (e.g. Streisinger et al., 1981; Naruse et al., 1985; Komen et al., 1993; Taniguchi et al., 1994; Young et al., 1996; Sarder et al., 1999). If doubled haploid mating designs are utilized, genetic analysis of crosses between lines have the benefits of ease of genetic typing and substantial phenotypic divergence among progeny (e.g., Robison et al., 2001; Nichols et al., 2004; Zimmerman et al., 2004). Doubled haploid designs appear to have substantially more experimental power than conventional designs for QTL detection (Martinez et al., 2005).

However, highly divergent mapping crosses are not the ideal populations in which to achieve MAS. Crosses of highly productive cultured strains that are suitable for use in breeding programs may not be highly divergent from each other for either marker loci or traits. Such crosses are the material in which MAS would likely be applied. If a set of candidate genes or QTL-associated marker loci can first be identified using divergent mapping crosses, then the marker loci could be utilized in the breeding crosses to test for hypothesized associations. If the associations are promising, the markers could be used to facilitate selection in the breeding crosses.

Ideally, it would be desirable to be able to extrapolate information about QTL locations from highly divergent experimental mapping populations to breeding populations, but our ability to do that successfully remains to be determined. The studies cited above indicate that there may be substantial conservation of QTLs across taxa, implying that QTLs identified in divergent mapping crosses might often be extrapolated successfully to breeding crosses. Testing for conservation of QTLs across a number of independent crosses within species should provide important information about the potential for streamlining the utilization of QTLs in breeding programs by allowing a restricted set of QTL marker loci to be utilized and tested in breeding crosses.

Acknowledgments

This work was funded in part by a U.S. Department of Agriculture Special Research Grant titled “Aquaculture, Idaho and Washington” to Gary Thorgaard and Ruth Phillips.

References

Albertson R.C., Streelman J.T. and T.D. Kocher, 2003. Directional selection has shaped the oral jaws of Lake Malawi cichlid fishes. *Proc. Natl. Acad. Sci. USA,* 100:5252-5257.

Andersson L., 2001. Genetic dissection of phenotypic diversity in farm animals. *Nat. Rev. Genet.,* 2:130-138.

Ben Chaim A., Paran I., Grube R.C., Jahn M., van Wijk R. and J. Peleman, 2001. QTL mapping of fruit-related traits in pepper (*Capsicum annuum*). *Theor. Appl. Genet*., 102:1016-1028.

Burke J.M., Tang S., Knapp S.J. and L.H. Rieseberg, 2002. Genetic analysis of sunflower domestication. *Genetics*, 161:1257-1267.

Burt D.W. and P.M. Hocking, 2002. Mapping quantitative trait loci and identification of genes that control fatness in poultry. *Proc. Nutr. Soc.,* 61:441-446.

Chagne D., Brown G., Lalanne C., Madur D., Pot D., Neale D. and C. Plomion, 2003. Comparative genome and QTL mapping between maritime and loblolly pines. *Mol. Breed.,* 12:185-195.

Chardon F., Virlon B., Moreau L., Falque M., Joets J., Decousset L., Murigneu A. and A. Charcosset, 2004. Genetic architecture of flowering time in maize as inferred from quantitative trait loci meta-analysis and synteny conservation with the rice genome. *Genetics,* 168:2169-2185.

Chen H., Wang S., Xing Y., Xu C., Hayes P.M. and Q. Zhang, 2003. Comparative analyses of genomic locations and specificities of loci for quantitative resistance to *Pyricularia grisea* in rice and barley. *Proc. Natl. Acad. Sci. USA,* 100:2544-2549.

Clark M.S., 2003. Genomics and mapping of Teleostei (bony fish). *Comp. Func. Genom.,* 4:182-193.

Cnaani A., Hallerman E.M., Ron M., Weller J.I., Indelman M., Kashi Y., Gall G.A.E. and G. Hulata, 2003. Detection of a chromosomal region with two quantitative trait loci, affecting cold tolerance and fish size, in an F2 tilapia hybrid. *Aquaculture,* 223:117-128.

Cnaani A., Zilberman N., Tinman S. Hulata G. and M. Ron, 2004. Genome-scan for quantitative trait loci in an F2 tilapia hybrid. *Mol. Genet. Genom.,* 272:162-172.

Coimbra M.R.M., Kobayashi K., Koretsugu, S., Hasegawa O., Ohara, E., Ozaki A., Sakamoto T., Naruse K., Okamoto N., 2003. A genetic linkage map of the Japanese flounder, *Paralichthys olivaceus*. *Aquaculture,* 220:203-218.

Danzmann R.G., Cairney M., Davidson

W.S., Ferguson M.M., Gharbi K., Guyomard, R., Holm L.-E., Leder E., Okamoto N., Ozaki A., Rexroad C.E. III., Sakamoto T., Taggart J.B. and R.E. Woram, 2005. A comparative analysis of the rainbow trout genome with 2 other species of fish (Arctic charr and Atlantic salmon) within the tetraploid derivative Salmonidae family (subfamily: Salmoninae). *Genome,* 48:1037-1051.

Erickson D.L., Fenster C.B., Stenoien, H.K. and D. Price, 2004. Quantitative trait loci analyses and the study of evolutionary process. *Mol. Ecol*., 13:2505-2522.

Georges M., 1997. QTL mapping to QTL cloning: mice to the rescue. *Genome Res*., 7:663-665.

Gharbi K., Gautier A., Danzmann R.G., Gharbi S., Sakamoto T., Hoyheim B., Taggart J.B., Cairney M., Powell R., Krieg F., Okamoto N., Ferguson M.M., Holm L.-E. and R. Guyomard, 2006. A linkage map for brown trout (*Salmo trutta*): chromosome homeologies and comparative genome organization with other salmonid fish. *Genetics,* 172:2405-2419.

Groenen M.A.M., Cheng H., Bumstead N., Benkel B.F., Briles W.E., Burke T., Burt D.W., Crittenden LB., Dodgson J., Hillel J., Lamont S., Ponce de Leon A., Soller M., Takahashi H. and A. Vignal, 2000. A consensus linkage map of the chicken genome. *Genome Res.,* 10:137-147.

Kai W., Kikuchi K., Fujita M., Suetake H., Fujiwara A., Yoshiura Y., Ototake M., Ventkatesh B., Miyaki K. and Y. Suzuki, 2005. A genetic linkage map for the tiger pufferfish, *Takifugu rubripes*. *Genetics,* 171:227-238.

Kocher T.D., Lee W.J., Sobolewska H., Penman D. and B. McAndrew, 1998. A genetic linkage map of a cichlid fish, the tilapia, *Oreochromis niloticus*. *Genetics,* 148:1225-1232.

Komen J., Eding E.H., Bongers A.B.J. and C.J.J. Richter, 1993. Gynogenesis in common carp (*Cyprinus carpio*). 4. Growth, pheontypic variation and gonad differentiation in normal and methyltestosterone-treated homozygous clones and F_1 hybrids. *Aquaculture,* 111:271-280.

Leder E.H., Danzmann, R.G. and M.M. Ferguson, 2006. The candidate gene, *Clock*, localizes to a strong spawning time quantitative trait locus region in rainbow trout. *J. Hered*., 97:74-80.

Lin Y.-R., Schertz K.F. and A.H. Paterson, 1995. Comparative analysis of QTLs affecting plant height and maturity across the Poaceae, in reference to an interspecific Sorghum population. *Genetics,* 141:391-411.

Mackay T.F.C., 2001. The genetic architecture of quantitative traits. *Annu. Rev. Genet*., 35:303-339.

Marques C.M., Brondani R.P.V., Grattapaglia, D. and R. Sederoff, 2002. Conservation and synteny of SSR loci and QTLs for vegetative propagation in four *Eucalyptus* species. *Theor. Appl. Genet.,* 105:474-478.

Martinez V., Thorgaard G., Robison B. and M.J. Sillanpaa, 2005. An application of Bayesian QTL mapping to early development in double haploid lines of rainbow trout including environmental effects. *Genet. Res*., 86:209-221.

Martyniuk C.J., Perry G.M.L., Mogahadam H.K., Ferguson M.M. and R.G. Danzmann, 2003. The genetic architecture of correlations among growth-related traits and male age at maturation in rainbow trout. *J. Fish Biol.,* 63:746-764.

Mauricio R., 2001. Mapping quantitative trait loci in plants: uses and caveats for evolutionary biology. *Nat. Rev. Genet.,* 2:370-381.

Moen T., Hoyheim B., Munck H. and L. Gomez-Raya, 2004. A linkage map of Atlantic salmon (*Salmo salar*) reveals an uncommonly large difference in recombination rate between the sexes. *Anim. Genet.,* 35:81-92.

Naruse K., Ijiri K., Shima A. and N. Egami. 1985. The production of cloned fish in the medaka (*Oryzias latipes). J. Exp. Zool.*, 236:335-341.

Naruse K., Tanaka M., Mita K., Shima A., Postlethwait J. and H. Mitani, 2004. A medaka gene map: the trace of ancestral vertebrate proto-chomosomes revealed by comparative gene mapping. *Genome Res.,* 14:820-828.

Nichols K.M., Young W.P, Danzmann R.G.,

Robison B.D., Rexroad C. III, Noakes M., Phillips R.B., Bentzen P., Spies I., Knudsen K., Allendorf F.W., Cunningham B.M., Brunelli J., Zhang H., Ristow S., Drew R., Brown K.H., Wheeler P.A. and G.H. Thorgaard, 2003. A consolidated linkage map for rainbow trout (*Oncorhynchus mykiss*). *Anim. Genet.,* 34:102-115.

Nichols, K.M., Wheeler P.A. and G.H. Thorgaard, 2004. Quantitative trait loci analyses for meristic traits in *Oncorhynchus mykiss*. *Environ. Biol. Fish.,* 69:317-331.

O'Malley K.G., Sakamoto T., Danzmann R.G. and M.M. Ferguson, 2002. Quantitative trait loci for spawning date and body weight in rainbow trout: testing for conserved effects across ancestrally duplicated chromosomes. *J. Hered.,* 94:273-284.

Ozaki A., Sakamoto T., Khoo S., Nakamura N., Coimbra M.R.M., Akutsu T. and N. Okamoto, 2001. Quantitative trait loci (QTLs) associated with resistance/susceptibility to infectious pancreatic necrosis virus (IPNV) in rainbow trout (*Oncorhynchus mykiss*). *Mol. Genet. Genom.,* 265:23-31.

Paterson A.H., Lin Y.-R., Li Z., Schertz K.F., Doebley J.F., Pinson S.R.M., Liu S.-C., Sanrsel J.W. and J.E. Irvine, 1995. Convergent domestication of cereal crops by independent mutations at corresponding genetic loci. *Science,* 269:1714-1718.

Peichel C.L., Nereng K.S., Ohgi K.A., Cole B.L., Colosimo P.F., Buerkle C.A., Schluter D. and D.M. Kingsley, 2001. The genetic architecture of divergence between threespine stickleback species. *Nature*, 414:901-905.

Perry G.M.L., Danzman R.G., Ferguson M.M. and J.P. Gibson, 2001. Quantitative trait loci for upper thermal tolerance in outbred strains of rainbow trout (*Oncorhynchus mykiss*). *Heredity,* 86:333-341.

Perry G.M.L., Sakamoto T., Ferguson M.M. and R.G. Danzmann, 2005. Sex-linked quantitative trait loci for thermotolerance and length in rainbow trout. *J. Hered*., 96:97-107.

Rao G.U., Ben Chaim A., Borovsky Y. and I. Paran, 2003. Mapping of yield-related QTLs in pepper in an interspecific cross. *Theor. Appl. Genet*., 106:1457-1466.

Reid D.P., Szanto A., Glebe, B., Danzmann R.G. and M.M. Ferguson, 2005. QTL for body weight and condition factor in Atlantic salmon (*Salmo salar*): comparative analyses with rainbow trout (*Oncorhynchus mykiss*) and Arctic charr (*Salvelinus alpinus*). *Heredity,* 94: 166-172.

Rexroad C.E. III, Rodriguez, M.F., Coulibaly I., Gharbi K., Danzmann R.G. DeKoning J., Phillips R. and Y. Palti, 2005. Comparative mapping of expressed sequence tags containing microsatellites in rainbow trout (*Oncorhynchus mykiss*). *BMC Genomics,* 6:54.

Robison B.D., Wheeler P. A., Sundin K., Sikka P. and G.H. Thorgaard, 2001. Composite interval mapping reveals a major locus influencing embryonic development rate in rainbow trout (*Oncorhynchus mykiss*). *J. Hered*., 92:16-22.

Sakamoto T., Danzmann R.G., Okamoto N., Ferguson M.M. and P.E. Ihssen, 1999. Linkage analysis of quantitative trait loci associated with spawning time in rainbow trout (*Oncorhynchus mykiss*). *Aquaculture,* 173:33-43.

Sakamoto T., Danzmann R.G., Gharbi K., Howard P., Ozaki A., Khoo S.K., Woram R.A., Okamoto N., Ferguson M.M., Holm L.-E., Guyomard R. and B. Hoyheim, 2000. A microsatellite linkage map of rainbow trout (*Oncorhynchus mykiss*) characterized by large sex-specific differences in recombination rates. *Genetics,* 155:1331-1345.

Sarder M.R.I., Penman D.J., Myers J.M. and B.J. McAndrew, 1999. Production and propagation of fully inbred clonal lines in the Nile tilapia (*Oreochromis niloticus* L.). *J. Exp. Zool.,* 284:675-685.

Shapiro M.D., Marks M.E., Peichel C.L., Blackman B.K., Nereng K.S., Jonsson B., Schluter D. and D.M. Kingsley, 2004. Genetic and developmental basis of evolutionary pelvic reduction in threespine sticklebacks. *Nature,* 428:717-723.

Streisinger G., Walker C., Dower N., Knauber D. and F. Singer, 1981. Production of clones of homozygous diploid zebra fish (*Brachydanio rerio*). *Nature,* 291:293-296.

Taniguchi N., Han H.S. and A. Tsujimura,

1994. Variation in some quantitative traits of clones produced by chromosome manipulation in ayu, *Plecoglossus altivelis*. *Aquaculture,* 120:53-60.

Wada Y., Akita T., Awara T., Furukawa T., Sugai N., Inage Y., Ishii K., Ito Y., Kobayashi E., Kusumoto H., Matsumoto T., Mikawa S., Miyake M., Murase A., Shimanuki S., Sugiyama T., Uchida Y., Yanai S. and H. Yasue, 2000. Quantitative trait loci (QTL) analysis in a Meishan x Gottingen cross population. *Anim. Genet.,* 31:376-384.

Waldbieser G.C., Bosworth B.G., Nonneman D.J. and W.R. Wolters, 2001. A microsatellite-based genetic linkage map for channel catfish, *Ictalurus punctatus*. *Genetics,* 158:727-734.

Wilson K., Li Y., Whan V., Lehnert S., Byrne K., Moore S., Pongsomboon S., Tassanakajon A., Rosenberg G., Ballment E., Fayazi Z., Swan J., Kenway M. and J. Benzie, 2002. Genetic mapping of the black tiger shrimp *Penaeus monodon* with amplified fragment length polymorphism. *Aquaculture,* 204:297-309.

Woods I.G., Kelly P.D., Chu F., Ngo-Hazelett P., Yan Y.L., Huang H., Postlethwait J.H. and W.S. Talbot, 2000. A comparative map of the zebrafish genome. *Genome Res.,* 10: 1903-1914.

Wright D., Nakamichi R., Krause J. and R.K. Butlin, 2006. QTL analysis of behavioral and morphological differentiation between wild and laboratory zebrafish (*Danio rerio*). *Behav. Genet.,* 36:271-284.

Young W.P., Wheeler P.A., Fields R.D. and G.H. Thorgaard, 1996. DNA fingerprinting confirms isogenicity of androgenetically-derived rainbow trout lines. *J. Hered*., 87:77-81.

Young W.P., Wheeler P.A., Coryell V.H., Keim P. and G.H. Thorgaard, 1998. A detailed linkage map of rainbow trout produced using doubled haploids. *Genetics,* 148:839-850.

Yu Z. and X. Guo, 2003. Genetic linkage map of the eastern oyster, *Crassostrea virginica* Gmelin. *Biol. Bull*., 204:327-338.

Zimdahl H., Kreitler T., Gosele C., Ganten D. and H. Hubner, 2002. Conserved synteny in rat and mouse for a blood pressure QTL on human chromosome 17. *Hypertension,* 39: 1050-1052.

Zimmerman A.M, Evenhuis J.P., Thorgaard G.H. and S.S. Ristow, 2004. A single major chromosomal region controls natural killer cell-like activity in rainbow trout. *Immunogenetics*, 55:825-835.