Ultraviolet light in the salmonid environment

by

Iñigo Novales Flamarique

B.Sc., McGill University, 1988
B.Sc., McGill University, 1990

A Thesis Submitted in Partial Fulfillment of the
Requirements for the Degree of

MASTER OF SCIENCE

in the Department of Biology

ACCEPTED
FACULTY OF GRADUATE STUDIES
[redacted]
DEAN
DATE 28 Apr 93

We accept this thesis as conforming
to the required standard

[redacted]

Dr. C.W. Hawryshyn, Supervisor (Dept. of Biology)

[redacted]

Dr. P.T. Gregory, Departmental Member (Dept. of Biology)

[redacted]

Dr. J.F.R. Gower, Outside Member (Institute of Ocean
Sciences, Sidney, Remote Sensing Unit)

[redacted]

Dr. C.R. Barnes, Outside Member (Center
for Earth and Ocean Sciences)

[redacted]

Dr. T.P. Quinn, External (School of Fisheries, University of
Washington, Seattle)

University of Victoria

Supervisor: Dr. Craig W. Hawryshyn

ABSTRACT

The spectral characteristics of fresh and coastal waters of Vancouver Island were examined using an underwater spectroradiometer. The results were analyzed with respect to salmonid vision. Special emphasis was placed on the characterization of the ultraviolet part of the radiant spectrum, from 300 to 450 nm, to test whether UV light intensities in nature were comparable to those used in laboratory experiments eliciting fish visual responses. The angular distribution of light observed in meso-eutrophic waters was approximated theoretically using Mie scattering theory. From these approximations, percent polarization of the scattered radiances measured at two angles were computed and compared to values required for salmonid orientation using polarized UV light. The results show that there was sufficient UV light for salmonid visual processes up to approximately 15 meters in both lakes and coastal waters. Similarly, salmonid orientation using UV polarized light was found to be possible near surface waters (less than 6 meters from the surface), but using mostly wavelengths from 390 to 450 nm. An analysis of visual responses showed that spectral sensitivity curves from processing of any light cue conformed accurately to non-linear curve fits.

[redacted]

Dr. C.W. Hawryshyn, Supervisor (Dept. of Biology)

[redacted]

Dr. P.T. Gregory, Departmental Member (Dept. of Biology)

[redacted]

Dr. J.F.R. Gower, Outside Member (Institute of Ocean
Sciences, Sidney, Remote Sensing Unit)

[redacted]

Dr. C.R. Barnes, Outside Member (Center
for Earth and Ocean Sciences)

[redacted]

Dr. T.P. Quinn, External (School of Fisheries, University of
Wahington, Seattle)

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ACKNOWLEDGEMENTS

This study could not have been completed without my dive partners (Andrew and Michael Hendry) and all the persons who operated the computer to gather the data (Craig W. Hawryshyn, Luc Beaudet and Michael Hendry). I also gained insight into the problems through discussions with my supervisor Craig W. Hawryshyn, and my colleagues and friends Luc "F" Beaudet, David "Doug" Coughlin and Craig “the Stick” McDonald, thanks a million guys!!. Howard Browman and Daryl Parkyn were very helpful editing the first and second manuscripts respectively. A special thanks is forwarded to my roomies and friends, the Victoria Gilbert and Sullivan Society, the Victoria Operatic Society, the University of Victoria Choir, the University of Victoria Jive and Ballroom Dance Club, my soccer buddies, and all the people who, in one way or another, prevented me from going insane while completing this thesis.

People are always blaming their circumstances
for what they are. I don't believe in circumstances.
The people who get on in this world are the
people who get up and look for the circumstances
they want, and, if they can't find them, make them.

Bernard Shaw

To all those who regard science
as just another hobby..

Chapter 1

General Introduction

The study of light transmission through water started with the application of Smoluchowski's (1908) and Einstein's (1910) scattering theory to explain laboratory optical results using pure water (Dawson and Hulburt, 1937). Since then physical oceanographers have used this theory, Rayleigh's theory (Rayleigh, 1884) and the more general Mie theory (Mie, 1908) to explain scattering of light by waters containing particulates (Jerlov, 1976). Experimental work measuring underwater light intensities to correlate with theoretical predictions has been restricted to the open ocean (Smith and Baker, 1979; McFarland and Munz, 1975), oligotrophic lakes (Smith et al., 1973) and surface waters of small lochs (less than 2m depth,Spence et al., 1971). As a consequence, the light field of more productive waters bearing non-homogeneous particulates, fluctuating phytoplankton communities and staining by runoff and plant pigments was, prior to this work, documented insufficiently (see however results for dystrophic lakes, Bowling and Tyler, 1986). In particular, the ultraviolet (UV) part of the spectrum with wavelengths below 400 nm had never been studied. It is mainly due to this lack of photic information in the UV range, and the presumed importance of UV light for visual processes in various species of juvenile fishes (cyprinids, Avery et al., 1983, yellow perch, Loew and Wahl, 1991, rainbow trout, Browman et al., 1992) that this project was undertaken.

The visual system of a fish, like most sensory systems in any animal, is shaped through evolution to confer the individual ecological advantages. In the case of juvenile salmonids, the discovery of a UV cone (Bowmaker and Kunz, 1987), i.e. a cone photoreceptor that can detect UV light, arouse many hypotheses regarding its function during the early ontogeny of the animal. Among these, better foraging performance, and orientation using polarized UV light, have been recently demonstrated in laboratory conditions (Browman et al., 1992; Hawryshyn et al., 1990). Other uses for UV light in communication and recognition of conspecifics have also been suggested (Hawryshyn, 1992), although evidence to date is lacking for these hypotheses (Denton and Nicol, 1965).

Having a sensory mechanism that can detect polarized light, and being able to use it for orientation purposes may have important implications during migration of fishes (Hawryshyn et al., 1990). Because the sun is always located approximately normal to the center of the toroid formed by the underwater polarized light field (Hecht and Zajac, 1974; Waterman, 1954), being able to see this polarized light pattern can give the animal valuable information on its location and course. Therefore this may be one of a number of sensory systems that salmon use during their migrations (Quinn and Dittman, 1990).

The main part of this project was to characterize the randomly polarized UV light field in fresh and coastal waters of Vancouver Island, and to relate the findings to salmonid vision. The objective was to assess whether UV vision could be relevant in nature. To accomplish this, we studied the waters of Lake Cowichan (chapter 2) and coastal areas (Saanich Inlet and Trevor Channel, chapter 3) to test whether the light levels that elicit salmonid responses during laboratory experiments (Browman and Hawryshyn, 1992, Beaudet et al., 1991) were also present in the natural environment. The procedure followed was to multiply the light irradiances (intensities) by transmission coefficients giving the percentage of light that would traverse the ocular media of young rainbow trout (Hawryshyn et al., 1989), and by the wavelength-dependent absorption coefficients of the pigments found in the juvenile fish (UV, short (blue), middle (green) and long (red) wavelength sensitive) (Bernard, 1987, pers. comm.). The first multiplication corrected for absorption by the lens, cornea and vitreous humour of the fish's eye, the second accounted for the differential absorption of each pigment as a function of the incident wavelength. The results thus represent the available irradiance to stimulate each type of cone receptor.

Although polarized light measurements were not taken during this project, we attempted to theoretically calculate polarized light intensities using Mie theory applied to particles from Lake Cowichan, the fresh water location studied. This section of the project, outlined in chapter 5, evaluates the possiblity of navigation using polarized light in meso-eutrophic waters. It is part of a study where the main objective was to understand the biophysics of light scattering, and to approximate theoretically the light irradiances observed (chapter 4).

The final chapter in this thesis is an attempt to model spectral sensitivity curves derived from light-adapted goldfish and rainbow trout. These curves are the result of complex neural interactions that take place at every level in the retina (see, for example, Hope and Dawson, 1979, DeMarco and Powers, 1991). Previos modeling was based on empirical approximations from the absorption spectra of the animal's photoreceptors (Yager, 1967, Newmeyer, 1984). Although these fits have been effective when simulating parts of spectral sensitivity curves, an equation that accurately fits visual data across the spectrum has never been proposed. We found such an equation in Sirovich and Abramov's (1977) general formula for the analysis of "pseudopigments" (which are the resulting curves from interactions of two or more pigments). These results are shown in chapter 6.

In summary, the objectives of this thesis were as follows:

1. To measure the spectral characteristics of Lake Cowichan and coastal waters of Vancouver Island (Saanich Inlet and Trevor Channel), and to relate the findings to salmonid vision. In particular the visual possibilities derived from the light measurements should be related with the visual system of young salmon (<30g), since it is at this stage that the entire retina contains UV cones (unpublished observations by the author for steelhead trout and Sockeye salmon). Thus, the results should be interpreted within the realm of in-lake, estuary, and seaward migration; although large rainbow trout, with a smaller number of UV cones, are also UV sensitive (Coughlin and Hawryshyn, In Press). If these results also apply to returning Pacific salmon, then the light measurements presented can also indicate navigation possibilities using polarized light for ocean dwelling salmon (e.g. migration between open ocean feeding grounds and home migration).

2. To gain an understanding of the physical mechanisms giving rise to the observed light fields by estimating the distribution of scattered light at specific angles based on Mie theory.
3. To predict theoretically the percent polarization of scattered light at various angles and to assess its use during salmonid migration, and
4. To model spectral sensitivity curves throughout the entire visual spectrum.

Chapter 2

The photic environment of a salmonid nursery lake

Introduction.

Recent discoveries showing that salmonids possess UV photoreceptors (brown trout, Salmo trutta, Bowmaker and Kunz, 1987) and that they are capable of detecting ultraviolet radiation (rainbow trout, Oncorhyncus mykiss Hawryshyn et al., 1989) have led to hypotheses regarding its use in nature. Among these hypotheses, contrast enhancement between target and background through UV vision has been suggested as a means of better foraging performance in larval fishes (Bowmaker and Kunz, 1987; Loew and McFarland, 1990). Also, UV vision and, in particular, detection of UV polarized light, may be a navigational tool used by juvenile salmonids to exit nursery lakes and coastal areas on their way to the open ocean (Hawryshyn et al, 1990). The spectral characteristics of these inshore and coastal environments, however, have not been fully examined.

The study of UV transmission in water bodies has concentrated on the open waters of oligotrophic oceans (Smith and Baker, 1979). Yet, the juvenile stage of some salmonids (i.e. sockeye salmon, Oncorhyncus nerka nerka, cutthroat trout, Oncorhyncus clarki, steelhead trout, Oncorhyncus mykiss) occurs in mesotrophic and/or eutrophic lakes. These water bodies usually contain high concentrations of chlorophyll and dissolved organic matter (DOM) which control the seasonal variations of light transmission in them (Prezelin et al., 1991). As such, the visual environment experienced by juvenile salmonids of some species is very different from what they encounter as adults in the open ocean, the spectral characteristics of the open ocean being highly dependent on the water molecules alone (Jerlov, 1976; Kirk, 1983). Nevertheless, it is during their early life history in nursery lakes that juvenile salmonids possess UV vision, a property which appears to be lost during development (Hawryshyn et al., 1989; Beaudet et al. 1991). Thus, it is important to examine the availability of UV light in nursery lake environments of these Pacific salmonid species.

The validity of hypotheses regarding the use of UV vision in nature depends on the confirmation of two observations: first, the light stimuli to which fish respond in laboratory experiments must match the intensity cues present in the environment; second, the presentation of visual cues alone during laboratory experiments simulating natural conditions should result in the predicted fish behaviour. So far, none of these conditions have been met for waters representative of salmonid nursery lakes. Thus, results from laboratory studies describing salmonid behaviour to light stimuli of defined energy and wavelengths (Hawryshyn et al., 1989; Hawryshyn et al., 1990) have not been compared to ecologically relevant light intensities present in the natural environment. It was therefore the goal of this study to characterize the spectra of available light in Lake Cowichan, a nursery lake for various species of Pacific salmonids. In the following study, the distribution of light from the various parts of the spectrum (UV, short, middle and long wavelengths) with respect to depth and direction of observation is examined, how the light field varies with time of day, and how these observations relate to salmonid visual capabilities and hypotheses concerning utility of UV vision in nature.

An underwater spectroradiometer was used to measure irradiance at several depths and at different locations in Lake Cowichan. Some measurements lasted periods of twenty-four consecutive hours to examine changes in the light field with time of day. In particular measurements of the spectral characteristics of Lake Cowichan during crepuscular periods were emphasized since, at these times, the light field is expected to shift towards shorter wavelengths (Rayleigh and Mie scattering, Van de Hulst, 1957). To assess the amount of light available for stimulation of salmonid photoreceptors, the spectral irradiance values obtained were corrected for absorption of the four pigments found in juvenile salmonid retinae (UV, short wavelength (blue), middle wavelength (green) and long wavelength (red)) and for ocular media transmission of rainbow trout, O. mykiss. This resulted in values representative of the intensities of light available for photoreception in salmonids. The set of values obtained, therefore, constitutes an estimate of the region in the water column that can be used for cone photoreception. The amount of UV available in nature for stimulation of salmonid photoreceptors was then compared to irradiance values used in the laboratory (Hawryshyn et al., 1989). This answered whether there is enough UV light penetrating Lake Cowichan to drive visually-mediated behaviours.

Materials and Methods.

The site of study was Lake Cowichan (Vancouver Island, lat: 48 ° 40-55' North, long: 124 ° 5-25' West). Seven sampling stations were established to compare regional differences in types of water that may arise from proximity to outflows, inflows or differing catchment areas (Fig. 2.1).

Spectral irradiance measurements were taken using an underwater LiCor spectroradiometer (LI-1800 UW model, LiCor instruments, Lincoln, Nebraska). This instrument consists of three major components: a filter wheel, a holographic monochromator and a silicon detector with related electronics. Light enters the LI-1800 through a standard cosine collector and is directed through the filter wheel before entering the monochromator. The filter wheel contains seven order-sorting filters which eliminate second order harmonics. The use of the seven filters also enhances stray light rejection by filtering out light that is not in the same spectral region as that being examined. This results in improved performance as compared to normal, single monochromator systems (Biggs, 1984). There is also a dark reference (zero reading) on the filter wheel that is substracted from any output from the detector. The data obtained thus contain minimal noise levels. The LI-1800 used has a holographic grating bandwidth of 4 nm; accurate measurements can therefore be obtained from 300 to 850 nm. The irradiance values obtained are in quantal units (photons/(m^2*sec*nm)), each scan collecting light over an angle of 180° (spectral irradiance).

Spectral irradiance measurements were taken by SCUBA divers submerging the LiCor spectroradiometer to 18 metres depth and bringing it towards the surface at 3 meter intervals. Four scans ranging across the spectrum from 300 to 850 nm were taken at each depth. The scans show spectral irradiance of downwelling, upwelling, and horizontal light in the sun's and antisun's directions at 5 nm intervals (Figs 2.2, 2.3). The spectroradiometer was connected to a portable computer situated at the surface and operated by the boat tender-researcher. This person communicated with the divers using a surface-to-diver communication system and controlled the execution of the scans through the computer. An air scan was taken at the end of each dive and used to correct the irradiance values found underwater for differences in weather conditions among stations. This was done by dividing the irradiance values at each depth by the irradiance at the surface (air scan). An ANOVA and Duncan's Paired Grouping Test were performed to detect differences in light intensity (spectral irradiance) among stations. Downwelling light extinction coefficients were calculated for each station using the formula (adapted from Tyler and Preisendorfer, 1962):

$$K(<Z_1+Z_2>/2) = (1/(Z_2-Z_1))\ln\ (H(Z_1)/H(Z_2)),$$

where

$K(<Z_1+Z_2>/2)$ is the extinction coefficient for light travelling from depth Z_1 to depth Z_2 (in m^{-1}), and,

$H(Z_1)$ and $H(Z_2)$ are the downwelling irradiances at depths Z_1 and Z_2 respectively.

Quantum integrations over the UV (300-400 nm), short (401-470 nm), middle (471-570 nm) and long wavelength (571-765 nm) parts of the spectrum were carried out to show differences in relative intensities at particular times during the day. The ranges for each type of light correspond to the parts of the spectrum where the probability of quantal catch by each of the corresponding pigments (absorption) is highest (Fig. 2.4). Relative intensities were obtained by dividing the total amount of a certain type of light (e.g. UV) by the total light in the entire spectrum (Figs. 2.8 2.9, 2.10).

We used an eighth order polynomial template for vertebrate cone absorption to correct the irradiance values measured for absorption of the four pigments found in juvenile salmonid retina (Bernard, 1987; pers. comm.). The polynomial template yielded normalized absorbance curves for each pigment as a function of the ratio of wavelength of maximum absorbance to wavelength (Fig. 2.4). Absorptance values were then generated using the equation

Absorptance=1-Transmission, where

$\log_{10}$(Transmission)=-Absorbance.

Thus, multiplication of the spectral irradiance values by these normalized absorptances corrected for the differential absorptance of each pigment over its active range. The resulting values, now corrected for pigment absorption, were multiplied by coefficients giving the percentage of light transmitted for each wavelength through the ocular media of small rainbow trout, O. mykiss (Hawryshyn et al., 1989). The modified irradiance values obtained after these two corrections represent the relative amounts of photons of each wavelength that are available to stimulate the cone photoreceptors in the fish retina. This we denote the "available spectral irradiance" (other authors may call this “photon catch”, however, this terminology was not chosen because it implies that all the photons will be caught, or processed, by the visual system, a situation which may not be the case). The values are relative because the actual absorption will depend, among other factors, on the relative numbers of each type of photoreceptor in the retinal region being illuminated. Quantum integrations over the full absorption spectrum of each pigment were then performed to obtain the total available spectral irradiance from each type of light at each depth and in each direction.

UV light is absorbed by the Schiff base linkage of the chromophores in salmonids (ß absorption bands, Fig. 2.4). However, the sensitivity of the response to UV polarized light differs for these ß absorption bands and UV cones (Hawryshyn and McFarland, 1987). Stimulation of the ß bands of the middle and long wavelength cone mechanisms is an independent process from that of the UV cone alpha band because these three types of cones are different physical entities in the retinal mosaic. Thus, neural processing of UV polarized light from two different cones, which differ in E-vector sensitivity, may constitute a system for the discrimination of UV polarized light intensity and plane of polarization. Because of their potential role in orientation of young salmonids (Hawryshyn et al., 1990), further integrations were performed restricting the limits to the absorption ranges of these ß bands (310 to 390 nm and 390 to 450 nm for the middle and long wavelength mechanisms respectively, see Fig. 2.4). The purpose of these integrations was to examine to what depths these secondary bands could be stimulated.

Water samples from different depths were collected at stations 1 and 2. These samples were treated for chlorophyll a, b and c. Pigment concentrations were obtained trichromatically using the procedure in Jeffrey and Humphrey (1975) for mixed phytoplankton communities. This procedure allowed the detection of possible relationships between phytoplankton concentrations at specific depths and changes in the spectral irradiance curves.

All the mathematical analyses were carried out using the Li-Cor spectroradiometer software and the SAS (version 5) statistical package.

Results.

The light characteristics at most locations in Lake Cowichan were similar (Table 2.1), the differences arising to a great extent from the variable catchment areas (see footnote in Table 2.1). The light extinction coefficients show that UV radiation is attenuated most (Table 2.2). With increasing depth the light field exhibits its maximum in the mid-wavelength spectrum (around 560 nm, see Fig. 2.3). The small standard errors associated with the extinction coefficients suggest that these trends in light transmission are representative of all the stations studied.

Analysis of water samples collected from stations 1 and 2 demonstrated chlorophyll a (Chla) to be generally the most abundant pigment for all depths (Table 2.3). An abrupt rise in the concentration of this pigment occurs between 9 and 12 meters in location 1.

The attenuation of downwelling irradiance with depth is shown in Figure 2.3. Similar curve shapes were found at all the other locations for each orientation. Spectral irradiance values were usually observed to decrease in the following order: downwelling light, horizontal light (sun's direction), horizontal light (antisun's direction) and upwelling light (Fig. 2.5). In one special circumstance, deviations from these results were obtained (Fig. 2.6). It can be observed in this figure that the last scan (which was taken 1.5 meters from the bottom) overlaps the previous one at various wavelengths. The bottom, situated at 8.5 meters, was composed of fine grained sand which is a highly reflective material (Chen and Nagaraja Rao, 1968). Such a substrate is also important in terms of fish vision because it can produce up to 40% polarized light when illuminated with diffuse light, and this may enhance target-background contrast (Loew and McFarland, 1990).

An analysis of irradiance curves with respect to direction of observation shows that light not only differs in intensity with direction, but also in spectral composition (Fig. 2.7). Near the surface short wavelengths constitute a major part of the light in the horizontal antisun's direction. In the horizontal sun's and downwelling directions the light field peaks in the middle to long wavelengths. Upwelling light is composed primarily of middle wavelength photons, which is also the case in all directions with increasing depth.

The spectral characteristics of Lake Cowichan during crepuscular periods exhibit an increase towards shorter wavelengths. The relative intensities of UV and short wavelength light peak prior to sunrise and immediately after sunset (Fig. 2.8). The opposite trend was observed for the long wavelengths. The middle wavelength part of the spectrum is stable for most of the day but exhibits relative intensity peaks of short duration during crepuscular periods. These peaks are the result of wavelenghts from 470 to 500 nm, which form the short wavelength part of the middle wavelength pigment integration spectrum. The spectral characteristics at dawn were examined with higher temporal resolution (Fig. 2.9). Although the curves follow previously mentioned trends, it can be further observed that the long wavelength part of the spectrum exhibits a local peak between, approximately, 4.6 and 6 a.m. The peaks in UV light during crepuscular periods can be observed more accurately by plotting the relative intensity of this type of light independently (Fig. 2.10).

Correction of the irradiance values for ocular media transmission and pigment absorption of rainbow trout results in the bar graphs shown in Figure 2.11. In laboratory experiments, fish are presented with light stimuli of specific wavelengths and the responses are assessed electrophysiologically (e.g. Beaudet et al., 1991) or behaviourally (e.g. Hawryshyn et al., 1989). After a period of background adaptation that isolates the sensitivity of the UV photoreceptor mechanism, salmonids have been shown to respond to light stimuli of the order of 10^{13} photons/m^2/sec (using 380 nm stimulus, Browman and Hawryshyn, 1992;

Hawryshyn et al., 1989). The middle and long wavelength photoreceptor mechanisms are the least sensitive and may require up to 100 times the previous intensities to be stimulated (Hawryshyn, 1991). Nevertheless, Figure 2.11 shows that, in Lake Cowichan, there is enough downwelling light to stimulate all the photoreceptor mechanisms at all depths studied. However, as is the case for the UV mechanism (Fig. 2.11), the ß band of the mid-wavelength mechanism appears to have a stimulation threshold depth at 18 meters (Fig. 2.12). These conclusions were reinforced by calculating the available light at 19 meters given a UV extinction coefficient of 0.65 m^{-1} (Table 2.1; 13.5 meters depth) and finding it insufficient in both cases. UV photoreceptor stimulation by upwelling light is restricted to depths less than 9 meters (the value at 15 meters is questionable due to the high standard error associated with it), whereas reception from sidewelling light is restricted to depths less than 18 meters. At all depths, the middle and long wavelength receptors are the most likely to be stimulated. It is important to realize that these and previous conclusions are based on results from statistical tests which pool data from different locations. Analysis of specific locations alone may yield significant results at greater depths.

Table 2.1. At each sampling location the spectral irradiance values at 3 and 6 meters were divided by the spectral irradiance at the surface. These values were then averaged to give the mean value at each station. Values with the same letter grouping are not significantly different (Duncan's Paired Grouping Test). It can be observed that most stations share similar water characteristics.

Station[1]	Mean	Grouping
5	0.25805	A
2	0.18010	B
6	0.13704	C
7	0.11251	C
4	0.10517	C
3	0.10374	C
1	0.05839	D

[1] Station 2 is located in a pristine area and the current flowing is quite strong. Station 5 is located near the outflow of the lake. These two characteristics (unaltered catchment area and outflow point) may explain the high light transmission observed for these stations, if we consider the lake to be a sink of nutrients and particulates (Wetzel, 1975). Station 3 is a shallow lake (8.5 meters depth) and the catchment area surrounding it has been extensively logged. Station 1 is bordered by resorts and the city's sewage pipe dumps its contents in these waters.

Table 2.2. Light extinction coefficients for the downwelling UV, short, middle and long wavelength parts of the spectrum, and for total spectral irradiance. The values, calculated for the various depths studied, represent averages from the seven stations. A dash line means that the values could not be computed as the original irradiance data were within the noise band of the spectroradiometer (only irradiance values equal to or bigger than 10^{12} photons/m^2/sec were considered).

Depth (m)	Light	Ext. coeff (m^{-1})	Std. Error(m^{-1})
1.5	UV	1.147	0.078
	Short	0.492	0.057
	Middle	0.276	0.068
	Long	0.537	0.095
	Total	0.487	0.077
4.5	UV	0.914	0.040
	Short	0.457	0.027
	Middle	0.238	0.036
	Long	0.356	0.032
	Total	0.306	0.032
7.5	UV	0.996	0.053
	Short	0.474	0.025
	Middle	0.309	0.026
	Long	0.376	0.029
	Total	0.339	0.027
10.5	UV	1.143	0.019
	Short	0.466	0.009
	Middle	0.281	0.016
	Long	0.364	0.047
	Total	0.297	0.023
13.5	UV	0.653	0.009
	Short	0.427	0.017
	Middle	0.234	0.033
	Long	0.326	0.015
	Total	0.306	0.038
16.5	UV	____	____
	Short	0.347	0.044
	Middle	0.186	0.026
	Long	0.272	0.018
	Total	0.203	0.032

Table 2.3. Chlorophyll values (in mg/ml) and standard errors for stations 1 and 2. All values were obtained from averages of two samples.

Depth(m)	Station	Chla	Std.Err	Chlb	Std.Err	Chlc	Std.Err
3	1	0.326	0.097	0.043	0.019	0.395	0.227
	2	0.335	0.129	0.773	0.302	0.491	0.849
6	1	0.816	0.225	0.048	0.024	0	0
	2	0.501	0.254	0.846	0.324	0.064	0.796
9	1	12.9	3.04	1.46	0.553	0.756	0.621
12	1	14.1	1.56	0	0	0	0
15	1	3.98	0.396	1.87	0.151	2.35	0.024
18	1	11.2	5.03	0.743	0.927	0.708	1.20

Figure 2.1. Map of Lake Cowichan showing the seven stations of study.

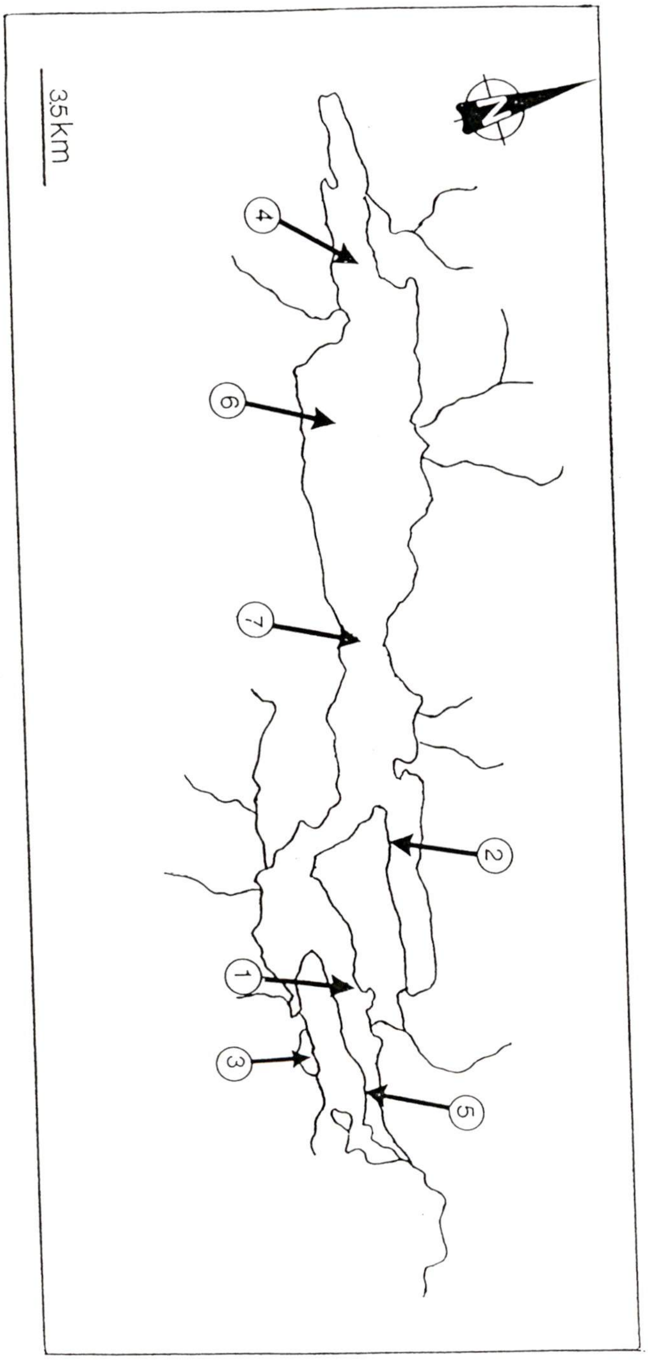


Figure 2.2. Spectroradiometer positions during scans. The sun's altitude is hypothetical; however, it is the altitude under which radiance measurements were taken in another study (see chapter 4; the angles are also used in that chapter for Mie calculations).

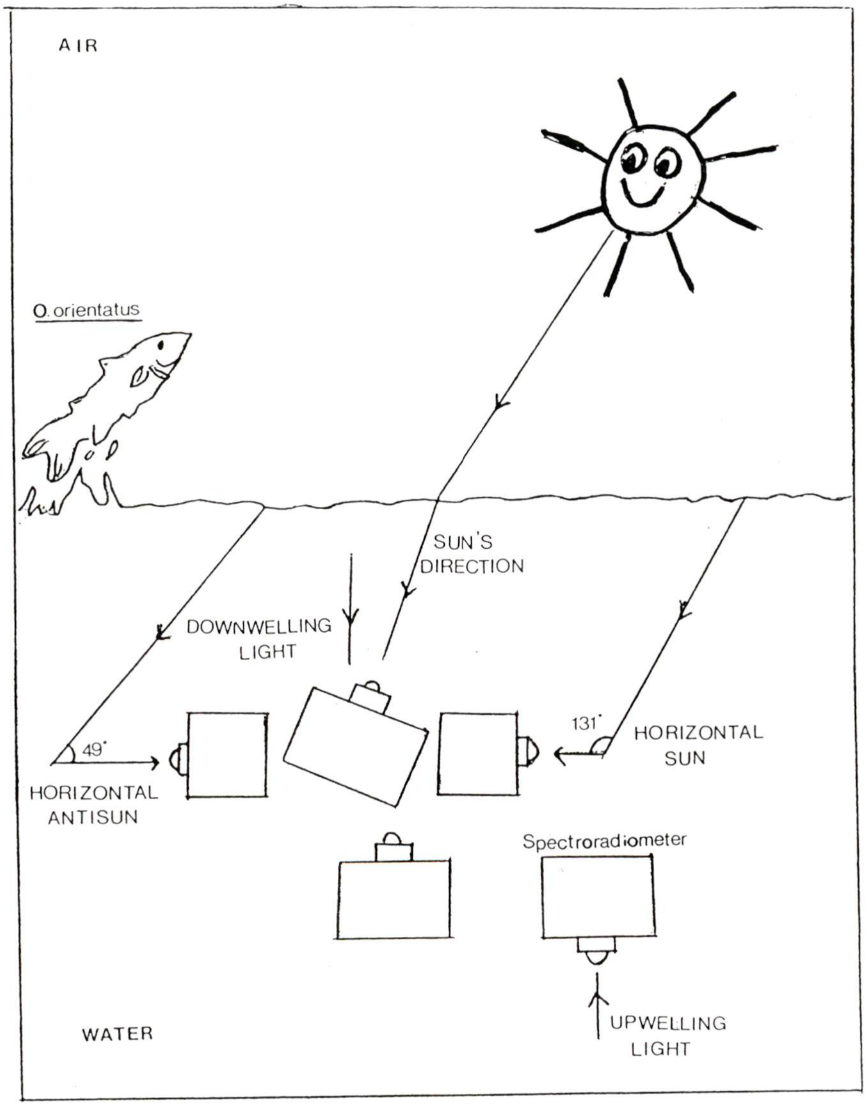


Figure 2.3. Downwelling spectral irradiance, at different depths, from 18 meters to the surface at location 1 (date: 27th of June 1991, time: 10:00-10:40 hours) . Each curve corresponds to one spectral scan. The scans were taken under clear sky conditions (sun's altitude: 39° (start) - 52° (end)). Mild winds created surface ripples which are responsible for the peaks in the scan taken at 0.3 meters. Similar trends with depth were also found under cloudy skies; however, in these cases the spectral irradiance values were lower and dependent on the thickness of cloud cover.

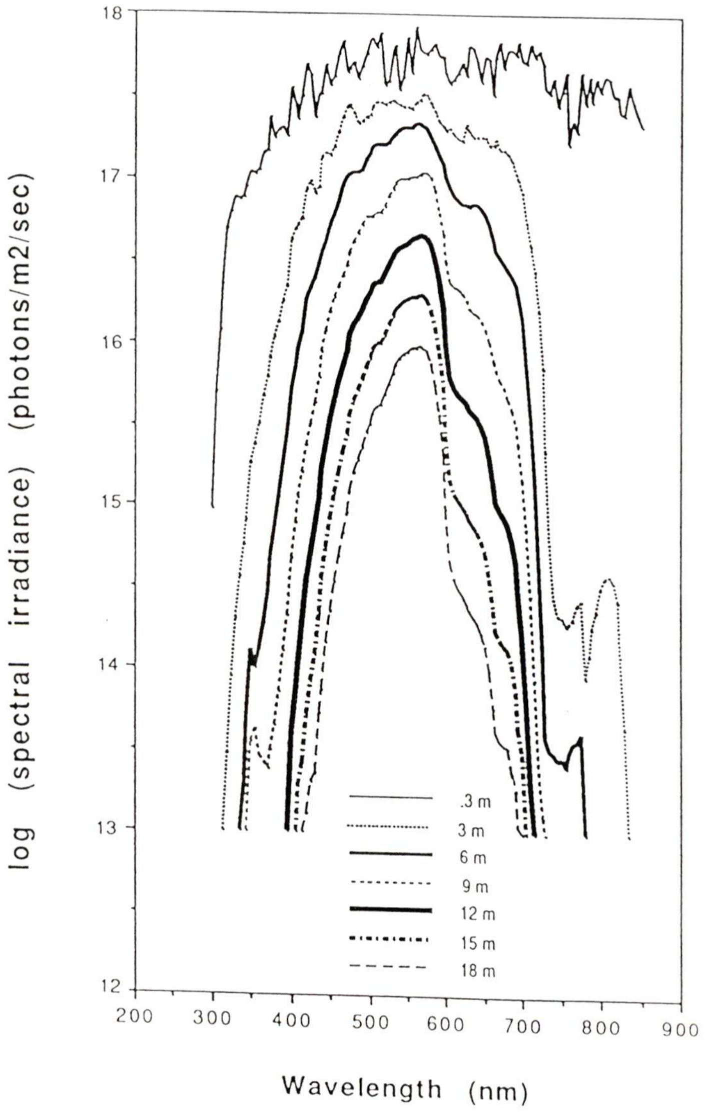


Figure 2.4. A plot of the normalized absorbance spectra of each pigment in juvenile salmonid retina (rainbow trout) as calculated from Bernard's (1987, pers. comm.) vertebrate cone template. Notice the ranges of maximum probability of quantal catch for the alpha absorption bands of each pigment (i.e. the range of wavelengths for which the main peak of a given pigment has the highest relative absorbance) and for the ß absorption bands of the middle and long wavelength pigments. These ranges define the integration limits for the four types of light studied (UV, short, middle an long wavelengths).

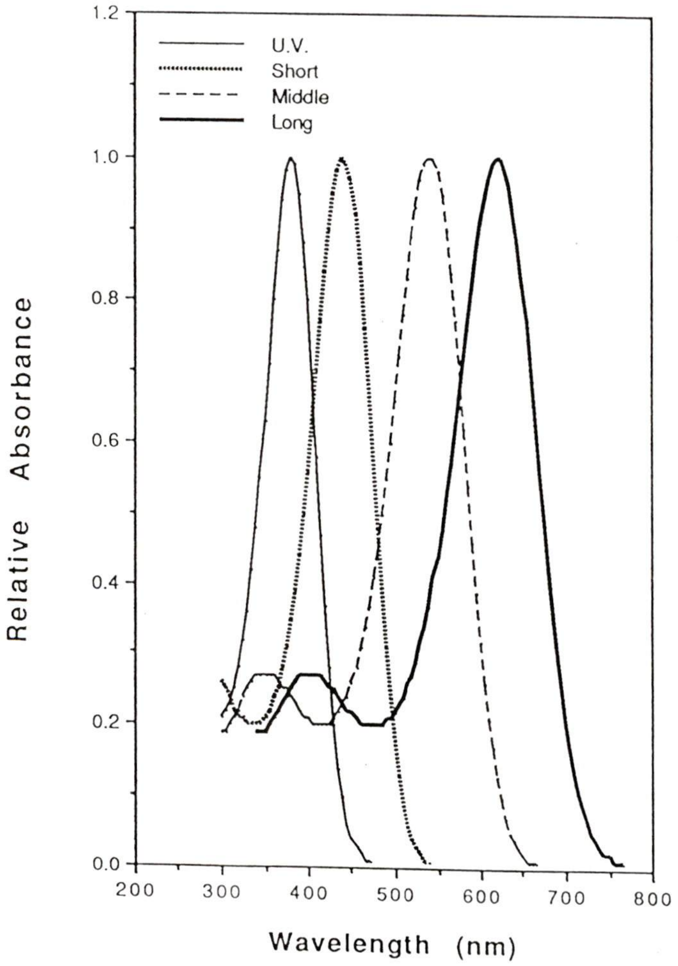


Figure 2.5. Differences in light intensity with direction for location 1, at 6 meters depth. Atmospheric conditions were the same as those in Figure 2.3. The same trends were detectable at all depths studied. However, with increasing depth, the differences became smaller and the light more monochromatic (see Figure 2.7).

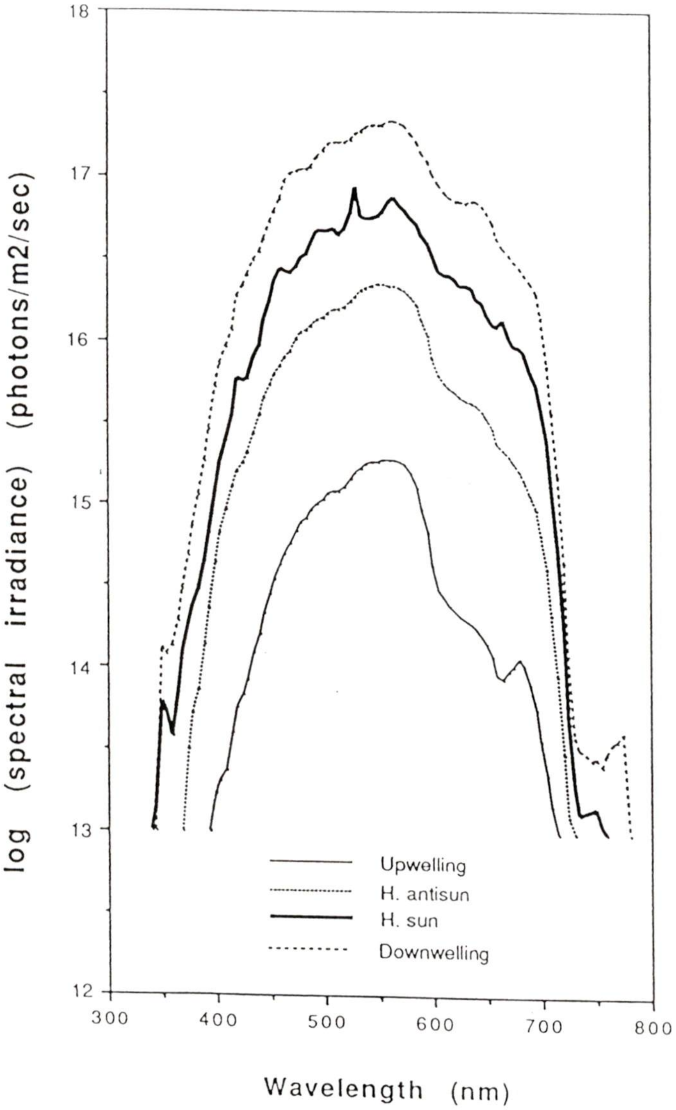


Figure 2.6. Downwelling spectral irradiance at station 3 (date: 19th of June, 1991, time: 11:04 hours). The scans were taken under clear sky conditions. Notice the increase in irradiance at 7 meters due to reflected light from the bottom of the lake.

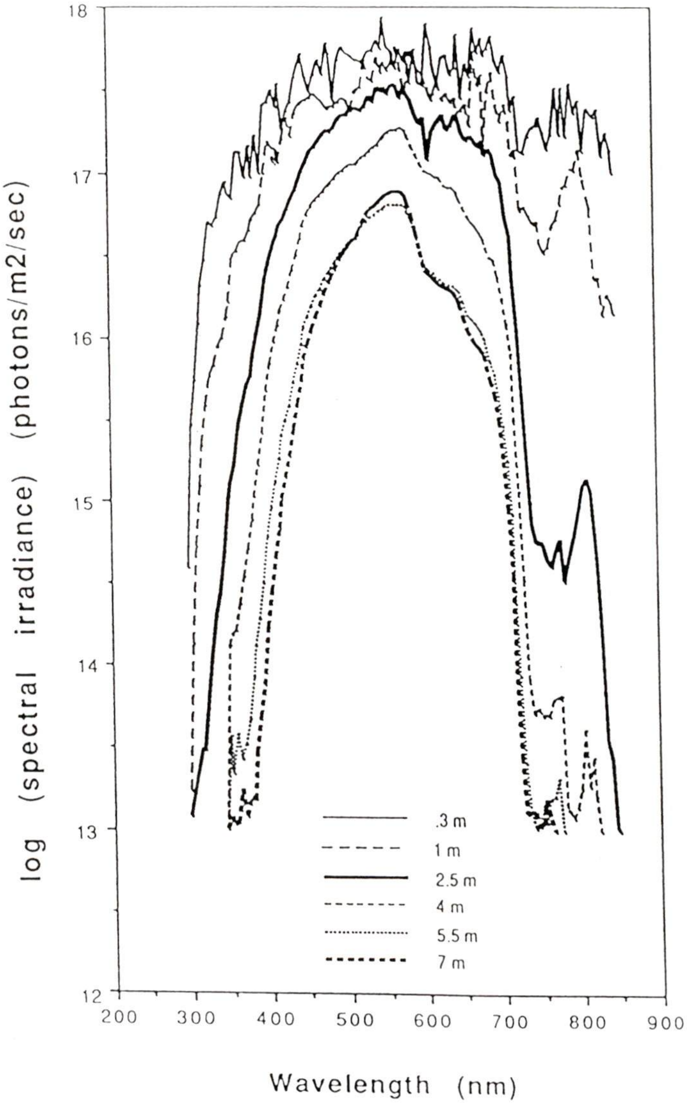


Figure 2.7. Log_{10} average spectral irradiance and associated standard errors (vertical bars) for each type of light from all stations (n=7). Direction abbreviations are as follows: UP: upwelling light, HA: horizontal antisun, HS: horizontal sun, and DO: downwelling light. Each depth plot is divided into the previous four orientations, and, within each orientation, bar graphs show the amounts of UV, short, middle and long wavelengths present in the water column. In surface waters, light in the horizontal antisun direction contains a large quantity of scattered short wavelength photons. However, downwelling and horizontal sun light is composed mostly of middle to long wavelength radiation. Upwelling light peaks in the middle wavelengths which is also the case in every direction with increasing depth.

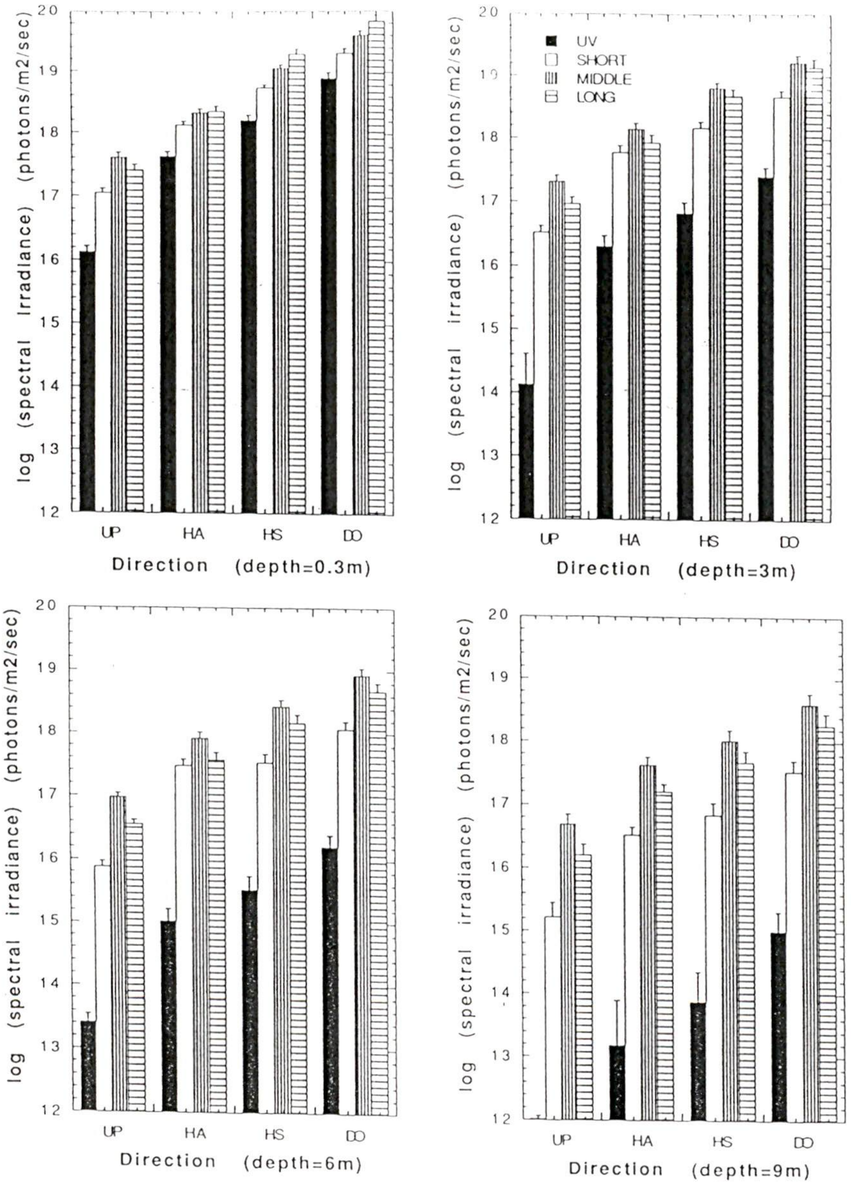


Figure 2.8. Relative intensity (irradiance) of each type of light to the total spectrum (300-850 nm) (date: 20th of June 1991). The downwelling scans from which the points derive were taken with the spectroradiometer lying at 3 meters depth facing the surface in station 1. The atmospheric conditions were complete overcast with irregular periods of rainfall. Shorter (UV and short) and middle wavelengths exhibit peaks during crepuscular periods and decrease in relative intensity during the day, the opposite trend can be observed for the long wavelength part of the spectrum. Long wavelength light exhibits local maxima at dawn and to a lesser extent at dusk. Sunrise and sunset occurred at 5:11 and 21:19 hours, Pacific Standard Time (Meteorological Information Center, Sidney, B.C.).

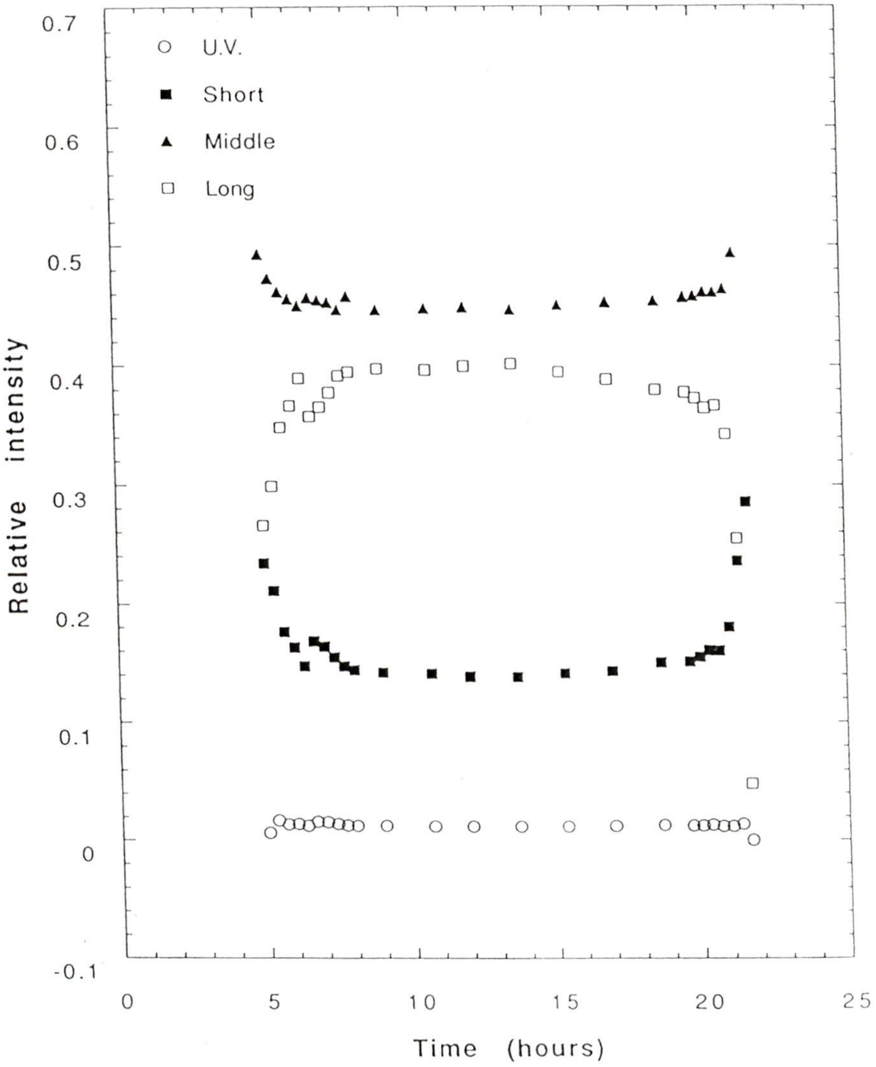


Figure 2.9. Spectral irradiance ratios of each type of light to the total spectrum during dawn (date: 28th of June 1991). Same collecting set-up as in Figure 2.8. Sunrise was predicted at 5:13 hours Pacific Standard Time (Meteorological Information Center, Sidney, B.C.). However local topography may have impeded the appearance of the sun until 6:60 hours explaining the rapid changes in slope observed in the curves at this time. The weather conditions were clear skies for the entire period. A local maximum in long wavelengths parallels the peaks from UV, short and middle wavelengths. This local peak is slightly displaced in time with respect to the shorter wavelength maxima as would be expected from the gradual change in pathlength of incoming solar radiation.

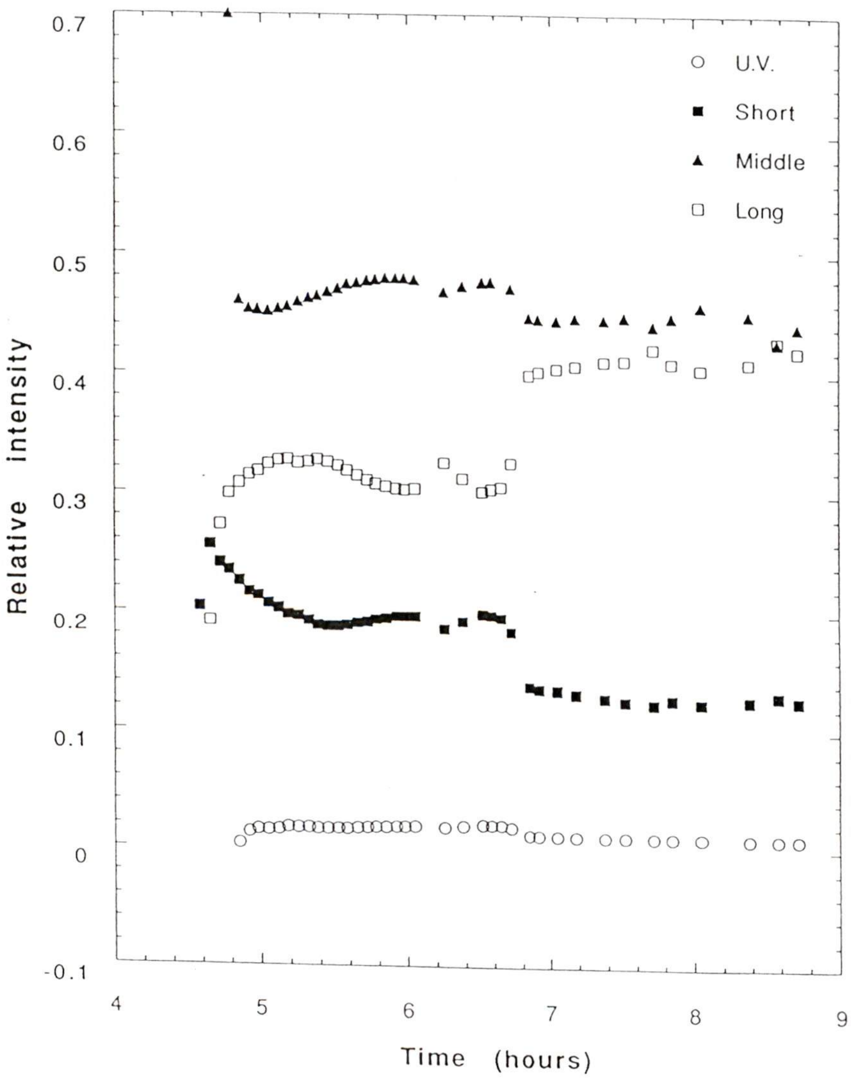


Figure 2.10. a) Relative intensity of UV light for an entire day, and, b) during dawn. Replotted from Figures 2.8 and 2.9.

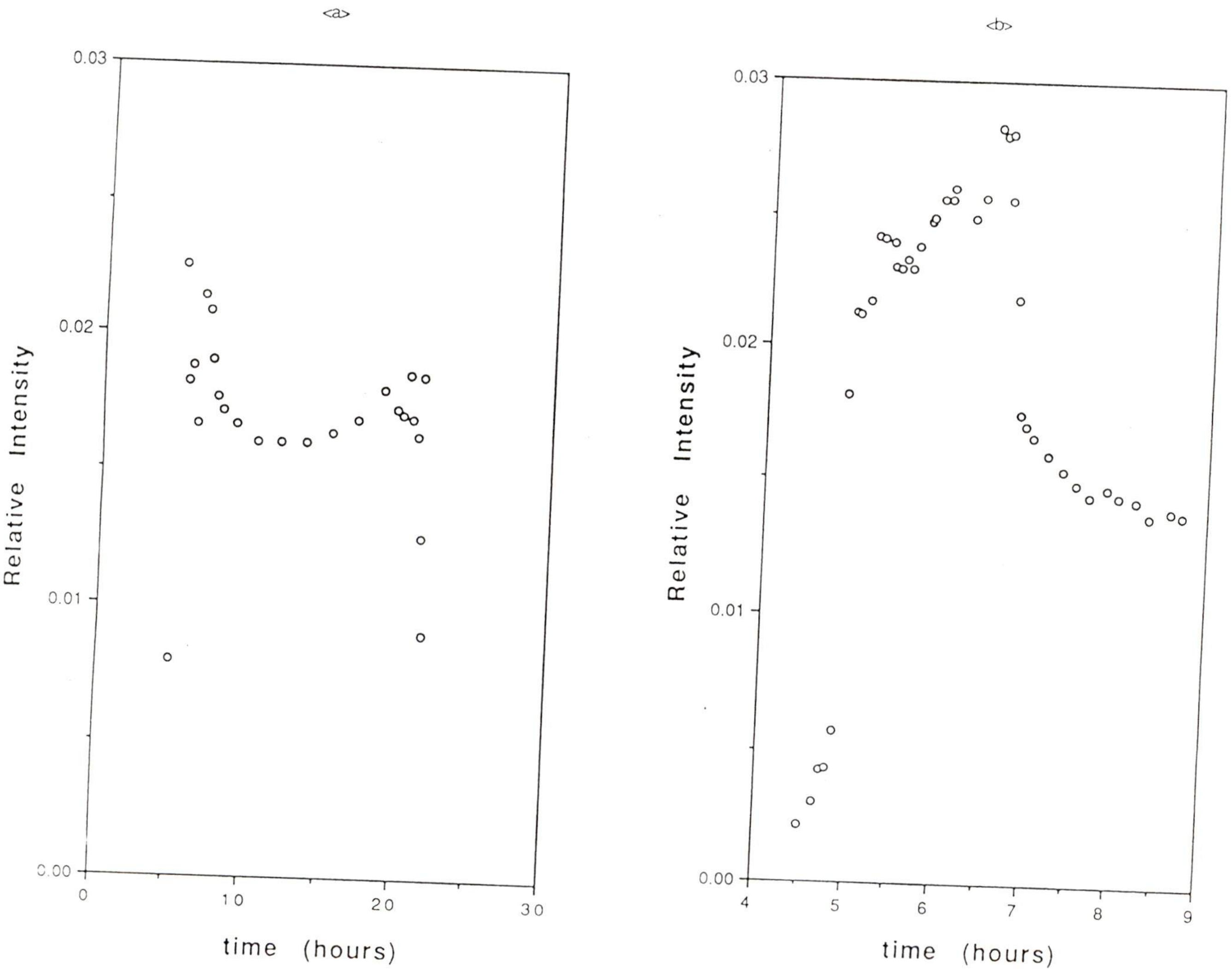


Figure 2.11. $\log_{10}$ average spectral irradiance values corrected for salmonid pigment absorption and ocular media transmission of small rainbow trout. Standard errors are indicated on the bar graphs (n=7). Presentation of data is the same as that in Fig. 2.7. These series of bar graphs represent a map of available light for activation of the different photoreceptor mechanisms with depth and direction.

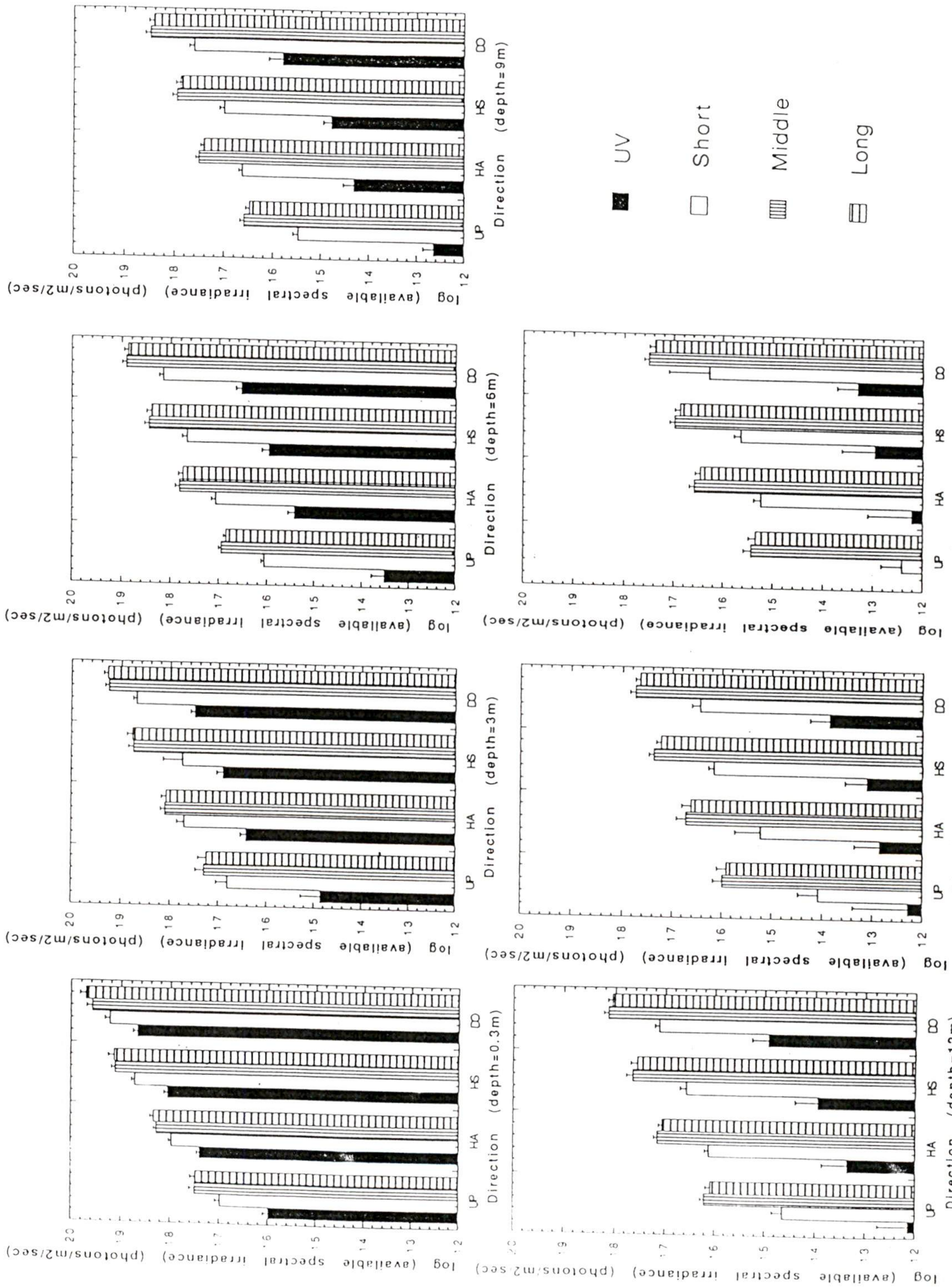
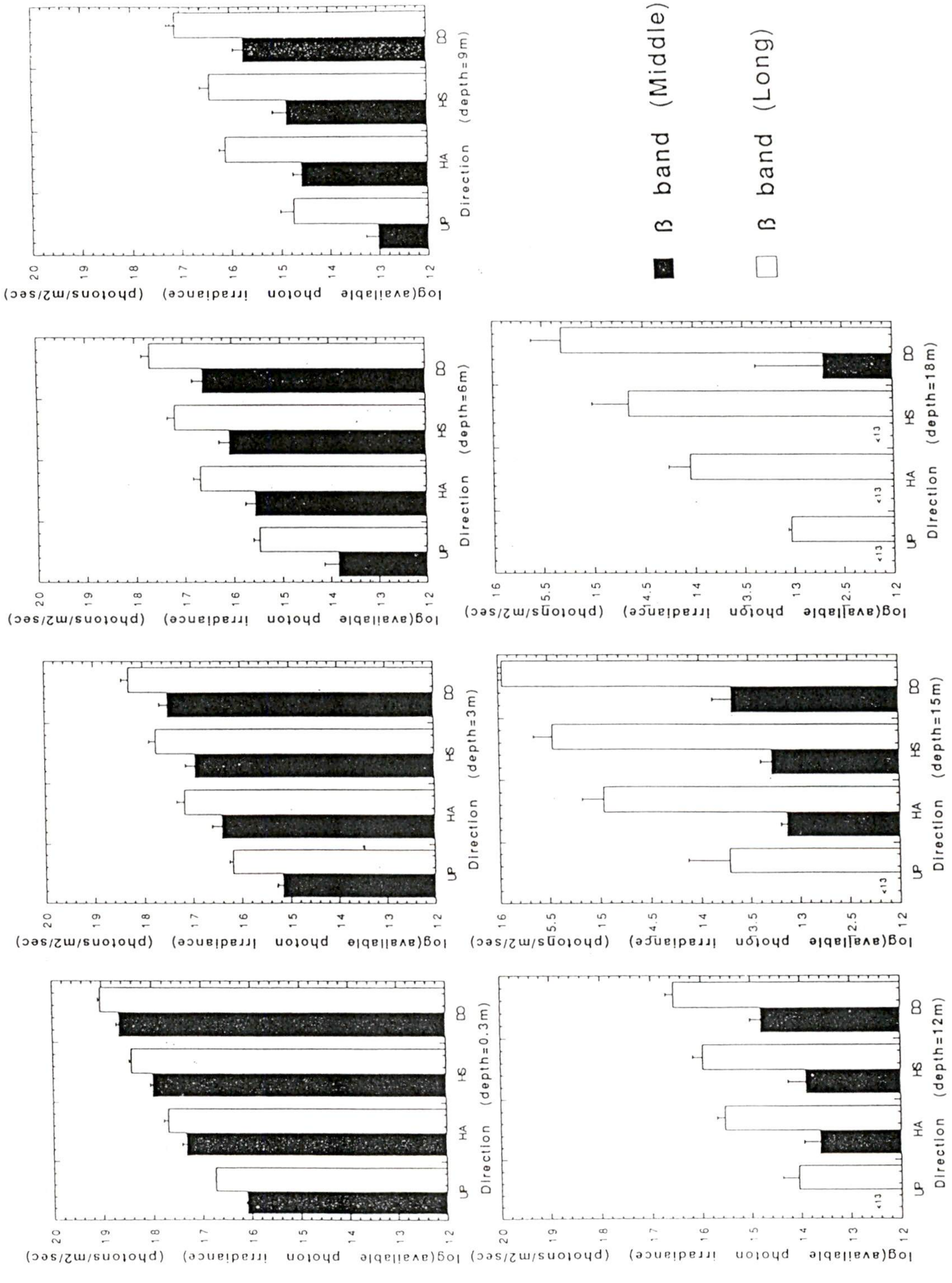


Figure 2.12. Log_{10} average spectral irradiance values corrected for the ß band pigment absorption curves of the middle and long wavelength cone mechanisms. Values were also corrected for ocular media transmission of small rainbow trout. Data presentation as in Figure 2.11.



Discussion.

The average extinction coefficients found for the various parts of the visual spectrum are at least five times those published for blue waters of the open ocean (Lenoble, 1954; Morel, 1965). Similarly, the extinction coefficients for total irradiance are much higher than those calculated for ultraoligotrophic lakes (Table 2.2; Smith et al., 1973). Lake Cowichan would be classified as a mesotrophic lake (Wetzel, 1975) or as having waters of type 1 (Jerlov, 1976). These discrepancies between lake Cowichan and blue waters of oligotrophic systems are essentially due to the higher concentrations of phytoplankton, DOM, organic and inorganic particulates in Lake Cowichan (see Heinerman and Ali, 1988 for lignin effects in mesotrophic lakes).

The tight link between phytoplankton populations and available light is exemplified by the high UV and blue extinction coefficients found at 10.5 meters (Table 2.2), and the presence of a bloom at this depth in location 1 (Table 2.3). Although a full spectrum of chlorophyll concentrations with depth is available only for location 1, these results are likely representative of the entire lake for two reasons: light penetration at the various locations is similar (Table 2.1) and the lake was stratified at the time of the study. The attenuation of middle wavelengths peaks around 7.5 meters and is probably the result of reflection from the first phytoplankton layer (located somewhere between 6 and 9 meters) (Davies-Colley et al., 1988). However, once in the phytoplankton layer (depth 10.5 meters), this type of light is the least attenuated. A possible explanation is that multiple scattering by phytoplankton cells is concentrating middle wavlelength light in the downwelling direction (Mie theory, Van de Hulst, 1957).

The differences in light transmission for the various parts of the spectrum can also be explained in terms of scattering theory (Jerlov, 1976). Small particles scatter most strongly the UV and short wavelengths while water molecules have absorption bands in the UV and long wavelength parts of the spectrum (Hecht and Zajac, 1974). Furthermore, any DOM present will absorb UV light extensively. Thus, in accordance with published results for the open ocean, the UV region of the spectrum was the most attenuated in our study. Attenuation of the short and long wavelength parts of the spectrum can be attributed mostly to absorption by the various chlorophylls (Wetzel, 1975).

Further analysis of the irradiance results shows that the light transmitted varies in magnitude and spectral composition depending on the direction, depth of observation and the time of day (see Jerlov, 1976 and McFarland and Munz, 1973). UV and short wavelengths are preferentially scattered by small particles and water molecules (Rayleigh, 1889). However, UV light is also absorbed more extensively than short wavelengths in the atmosphere and in the water column. As a result, the amount of UV light in the horizontal antisun direction is smaller than the amount of short wavelength light (Fig. 2.7). Downwelling light and horizontal light in the sun's direction exhibit maxima in the middle to long wavelength range. This is due to sunlight impinging directly on the cosine collector, longer wavelengths being scattered the least through the atmosphere. Upwelling light is maximum in the middle wavelengths as a result of scattering from phytoplankton and algal substrates.

The presence of backgrounds differing in hue with direction of observation has important consequences for the perception and tracking of objects in the water column. For instance, detection of dark objects is usually favoured against a bright background. Thus, a fish's position that would maximize luminance in the direction of observation (i.e. towards the sun) would be most beneficial. However, conspicuousness of lighter objects is often increased on a dark background, in which case vision in the antisun direction would be more effective. Furthermore, it has been suggested that reflected polarized light from fish scales may also be used by other fish to detect targets on dark backgrounds (a diffusing background producing less polarized light (Denton and Nicol, 1965)). These different scenarios suggest that an ideal orientation for maximum perception of light cues in surface waters is dependent on visual task. Fish may compromise between the previous advantages by travelling in the direction of the sun. In this case spectral irradiance in the forward direction is high yet some of the photons originating from sideways particle scattering would be polarized (and this, importantly, as the background becomes dimmer). Whether such preferred directions of fish travel related to background light cues exist is yet to be examined.

The gradient of light attenuation from surface to deeper waters also implies adaptability of the retina to varying spectral backgrounds (Singarajah and Harosi, 1992). As the fish travels down the water column, perception of a silhouette on a bright background becomes increasingly difficult (Le Grand, 1939). In deep waters, a point is reached when the light field is everywhere homogeneous (Jerlov, 1976) and the retinal sensitivity no longer varies with direction of observation. A highly reflective bottom in shallow waters creates an effect opposite to the attenuation mechanism just described. Because of reflection, upwelling light near the bottom can be more intense than, or nearly as intense as, that at lower depths (Fig. 2.6). Bottom predators may take advantage of the additional background illuminance and possible creation of polarized light (Chen and Nagaraja Rao, 1968) for target detection. This is perhaps another reason why darker animals may not be found near sandy bottoms, even at considerable depths.

Although the spectral composition at twilight has been examined by McFarland and Munz (1975b), these authors did not examine the UV part of the spectrum. Furthermore, a rigorous mathematical analysis for the various parts of the spectrum through a 24 hour period is not available. Results for Lake Cowichan for this period of time show opposite trends for the UV, short and middle wavelengths versus the long wavelength part of the spectrum (Figs. 2.9 and 2.10). Relative quantities of UV, short and middle wavelengths are most abundant during crepuscular periods, whereas relative amounts of long wavelengths increase during daylight hours. The long wavelength part of the spectrum also exhibits a local peak during crepuscular periods (Fig. 2.9). Some of these changes have been observed previously for the spectrum from 400 to 700 nm (McFarland and Munz, 1975b). They are due to preferential scattering of shorter wavelengths in the atmosphere by Rayleigh-size particles and increased scattering of longer wavelengths at particular pathlengths of incoming solar radiation (Lythgoe, 1979).

The relative increase in UV light during crepuscular periods may be important for salmonid navigation because these fish have been found to orient to the E-vector of polarized UV light (Hawryshyn et al., 1990). Given the relation between the sun's position and the most intense band of polarized light (approximately 90 degrees separation, Waterman, 1954), salmonids could potentially be using polarized light vision as an orientation mechanism during migration (Groot, 1965; Dill, 1971; Quinn, 1980). Interestingly, various authors have reported migratory episodes to occur during crepuscular periods (Johnson and Groot, 1963; Groot, 1965). Furthermore, the fish are observed to travel near the surface where stimulation of the UV cones is possible (Fig. 2.11).

The light patterns at dawn and dusk may also play a role in foraging if, indeed, UV vision enhances target-background contrast. The results for Lake Cowichan suggest that predators and prey with UV vision would be the most effective at obtaining food and lowering the risks of being captured respectively. In the case of juvenile salmonids, UV vision may be used to detect zooplankton as these organisms absorb high quantities of UV light (as suggested by Bowmaker and Kunz, 1987).

Another characteristic of the light field during crepuscular periods is its rapid changes in spectral composition. Two peaks of UV and short wavelengths occur before and at the moment when the sun rises over the horizon (Figs. 2.9 and 2.10b). These peaks are accompanied by local maxima in middle and long wavelength light, although these trends vary in time as well (Fig. 2.9, hours 5 to 7 a.m.). Camouflage of any sort could be very difficult in such a varying light field and thus this may be the cause for the observed high predation during crepuscular periods (Hobson, 1972). To counter these rapid changes in illumination, some coral reef fishes have developed combinations of pigments whose maximum absorptions approximate the peak wavelengths found in the environment during these time periods (Munz and McFarland, 1973).

The results from this study demonstrate that UV mediated visual behaviours are possible at all depths studied (Fig. 2.11). However, 18 meters was found to be the threshold depth for UV cone stimulation (Fig. 2.11). Such a depth restriction would force the fish to rise in the water column in order to profit from UV light cues for purposes of orientation. Surfacing, however, involves higher risks of predator detection from below and from outside the water column. A compromise could then be reached by swimming in a sinusoidal pattern, surfacing at regular intervals. Coincidentally, such a swimming pattern has been observed during salmonid migratory episodes (Groot, 1972; Westerberg, 1982; Ruggerone et al., 1990).

Chapter 3

Spectral characteristics of salmonid migratory routes from southern Vancouver Island (British Columbia)

Introduction

Although many researchers have examined the visual capabilities of fishes (Tomita et al., 1967; Yager, 1969; Bowmaker and Kunz, 1987; Hawryshyn and Harosi, 1991), few relate their findings to measured photic conditions in the environment (McFarland and Munz, 1975a,b; Muntz and Wainwright, 1978; Heinerman and Ali, 1985, 1989). Even in studies in which an attempt has been made at associating the visual capabilities of the animal with the light cues present in nature, these have sometimes been approximated because of lack of field measurements (Singarajah and Harosi, 1992). Spectral data are particularly lacking for coastal oceanic waters where river discharge and upwellings cause high levels of turbidity. The light levels in such environments differ markedly from those found in the open ocean (Jerlov, 1976). Nevertheless, young and adult salmonids must navigate through coastal waters to find their way to and from the open ocean.

The first goal in this study was therefore to measure the light levels in coastal waters of Vancouver Island under different atmospheric conditions (overcast vs. sunny), and to compare them with light intensities used in laboratory experiments eliciting fish responses (Beaudet et al., 1991). To test whether light intensity levels varied with distance from spawning rivers, measurements were taken along transects covering migratory trajectories along the coast. If a trend in UV light levels was found, salmonids could be using it to guide their migration (instead of, or along with, polarized light detection).

The second goal was to test the dependence of available light on water chemistry. In the case of UV light, DOM and Chla concentrations are the two major biophysical factors affecting its transmission (Prezelin et al., 1991), the light intensities were therefore correlated with these two factors. Because similar measurements were obtained for Lake Cowichan, the results in coastal and fresh water environments were compared to detect any photic changes that salmon may experience during their migrations. Such changes can be correlated with ontogenetic alterations in the retinal pigments found in some salmonids (Beatty, 1966), which may suggest differences in their visual ecology.

Materials and Methods

Spectral irradiance measurements from 18 m to the surface at 3 m intervals were collected at seven locations in Saanich Inlet (East coast of Vancouver Island) from July 9th to 26th, 1991, and at four locations in Trevor Channel (West Coast of Vancouver Island), from August 22nd to 31st, 1991 (Fig. 3.1). These locations are on two major migratory routes for Pacific salmonids spawning in Vancouver island streams.

The collection of data was accomplished by SCUBA divers as in the Lake Cowichan study. Correction of data to estimate available irradiance for stimulation of the fish's photoreceptors was as described in chapter 1.

It should be noted that in the previous calculations the relative absorbances per wavelength as given by Bernard's (1987) polynomial template equation were used to calculate absorptance. Thus, the available spectral irradiances values are dependent on the absolute absorbance of the pigment at its wavelength of maximum absorption. In order to calculate true absolute absorptance (as opposed to the relative absorbance-dependent values presented here and in the first chapter), one must multiply the relative absorbance per wavelength (e.g. from Bernard, 1987) by the total absolute absorbance (or optical density) of a photoreceptor along its axis (Harosi, 1975). Using published optical density values for amphibians and fish (average value used: 0.0124/µm, Harosi, 1975) and mutliplying by the average cone outer segment length of rainbow trout (10µm, Harosi, pers. comm.), we arrived at a total absorbance of 0.124 (the outer segment of the cones contains the absorbing pigment). The relative absorbance values generated from Bernard's (1987) polynomial template were multiplied by this value and the true absolute absorptances calculated using the previous formulae. Comparison of these quantities with previous relative-absorbance dependent values showed a maximum 9.3 fold difference when the relative absorbance was 0.01; however, values for the most absorbed wavelengths differed 3 times (for the wavelength of maximum absorbance) to 6.7 times (when the relative absorbance = 0.02) with previous calculations. Available spectral irradiance calculations using these new values for absorptance do not change the threshold depths for photoreceptor excitation and therefore the conclusions of this manuscript. The non-significance of the change is due to: 1) absorption factors (ranging from 0.002 to 0.24) are multiplied by very large numbers (spectral irradiances > 10^{13} photons/m^2/sec), 2) absorptance results obtained from absolute absorbance values for the highest absorbed wavelengths differ the least with relative absorbance generated results (thus altering little the overall integration results), and 3) the small alterations are within the standard errors for the various types of light at threshold depths.

To detect trends in spectral irradiance with distance from freshwater spawning rivers, measurements were taken along paths followed by salmonids spawning at Goldstream Provincial Park and Cowichan River (Fig. 3.1). Downwelling light measurements at each site were obtained by lowering the spectroradiometer from the side of the boat and holding it at 3 meters depth. Water samples were collected and tested for Chla concentrations and particle counts as described in chapter 1.

To determine whether different weather conditions changed the intensity of light available for salmonid vision, scans at the various locations were taken under clear and cloudy (completely overcast) skies. Since the chemistry of water also plays a major role in the transmission of light through water, Chla analysis of water samples from the various depths was performed on two replicates. An estimate of the relative amounts of particulate matter at the various locations was also obtained using a Coulter Counter (model TA II, Coulter Electronics Inc., Hialeah, Florida). Using a 70 μm aperture tube, particles with radii from 0.5 to 12.7 μm were counted in 0.05 ml samples. In addition, DOM estimates were obtained from published Department of Fisheries and Oceans reports (Brown and Borstad, 1981). This information permitted us to correlate spectral irradiance with biophysical parameters in the environment.

Because similar measurements have been presented for Lake Cowichan in the first chapter, comparisons were possible not only among coastal regions but also with this lake. These comparisons may suggest environmental reasons for the varying rhodopsin/porphyropsin ratios in the retina of anadromous migrating fish at different times of the year (Beatty, 1966).

Mathematical analysis of results used SAS (version 5) and LiCor software.

Results

Spectral measurements

In laboratory experiments, the minimum UV light irradiances required for stimulation of spectrally isolated UV cones using 360 to 380 nm stimulus approximates 10^{13} photons/m^2/sec (Hawryshyn et al., 1989); this value becomes $6.1*10^{12}$ photons/m^2/sec after correction for pigment absorption and transmission through the ocular media. Sensitivity of the fish to other wavelengths may decrease two orders of magnitude in the middle to long wavelengths (Hawryshyn, 1991). Correspondingly, the intensities required to stimulate the middle and long wavelength cone mechanisms may be up to 100 times those required for the UV or blue cone mechanisms. In the case of UV photoreception, the minimum light levels required, after pigment and ocular media corrections, were found at 20 m depth in Trevor Channel (Fig. 3.2). In Saanich Inlet, this depth was 15 m (Fig. 3.3).

The intensity and spectral composition of the available irradiance to stimulate salmonid photoreceptors changes with depth and direction of observation (Fig. 3.3). In Saanich Inlet, long wavelengths dominate the spectrum in surface waters (< 6 meters depth), but, with increasing depth, the "visual" spectrum peaks in middle wavelengths. Light levels decrease in the following manner: downwelling, horizontal sun, horizontal antisun and upwelling (Fig. 3.4). This pattern is altered under cloudy conditions because the spectral irradiance differences between horizontal sun and antisun directions tend to vanish. However, such overcast trends depend on the thickness and constituency of the cloud cover (Waterman, 1956).

Directionality of the sun's rays under completely overcast skies was observed at up to 12 meters depth under some types of cloud cover (Fig. 3.5). Whether in the presence of clear or cloudy skies, stimulation of the UV photoreceptors in Saanich Inlet is only possible at depths less than 15 meters (Fig. 3.3), this is also the case for stimulation of the ß absorption bands of the middle wavelength photoreceptors (Fig. 3.6). The relative sensitivities of the ß bands being approximately 0.316 to 0.63 times those of the corresponding alpha bands (Beauchamp and Lovasik, 1973).

The downwelling spectral irradiances for Lake Cowichan, Saanich Inlet and Trevor Channel show that Lake Cowichan has, in general, the highest UV levels (Fig. 3.7). The rapid decrease in UV light levels in Saanich Inlet below 12 meters depth coincides with higher Chla concentrations compared to the other study locations (Fig. 3.8a). Correspondingly, middle wavelengths remain highest at all depths for Saanich Inlet since they are absorbed minimally by phytoplankton.

Water chemistry

A comparison between the averaged Chla concentrations in Saanich Inlet and those observed in Trevor Channel showed that both of these coastal waters had more Chla during the summer than Lake Cowichan (Fig. 3.8a; Brown and Borstard, 1981). Furthermore, the first phytoplankton layer was closer to the surface for both coastal waters. Trevor Channel exhibited higher levels of Chla and DOM near the surface than Saanich Inlet (Fig. 3.8c, Brown and Borstard, 1981).

The number of particles between 0.5 and 12.7 μm in radius was higher in Lake Cowichan than in Saanich Inlet (Fig. 3.8b). Such small-radii particle differences may indicate the relative amounts of polarized light in both environments (Rayleigh, 1889). In coastal waters, we observed large numbers of whitish "filament-like" aggregates which would dissociate upon touch and could therefore not be included in the particle counts. However, their surface area, in the order of centimeters, was sufficient to intercept large amounts of radiation.

The number of particles from the various stations in the Goldstream transect appeared, in general, to decrease with increasing Chla concentrations (Fig. 3.9). However, there was no significant relation between intensity of UV light and concentrations of Chla along both transects.

Figure 3.1. Study sites in Saanich Inlet and Trevor Channel (Lake Cowichan also shown). GS and CT black dots represent stations along Goldstream and Cowichan Bay transects. Black squares are the stations for which complete sets of spectral irradiance data with depth were obtained.

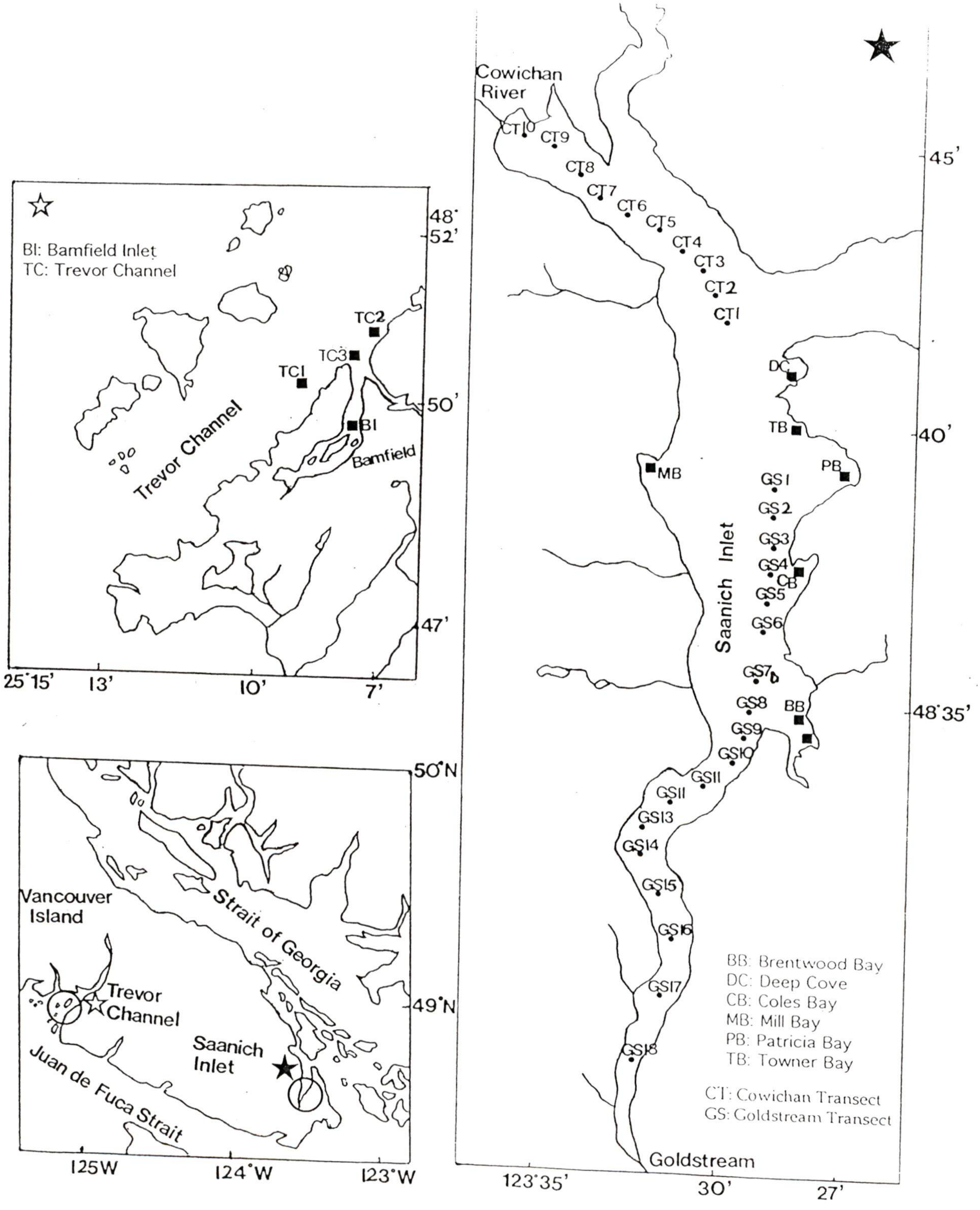


Figure 3.2. Spectral irradiance profile for Trevor Channel between 13:05 and 13:30 hrs on the 24th of August 1991 (TC1 station). Scans were taken under slightly overcast conditions (the sun's exposure was occassional and was otherwise visible through the cloud layers, sun's altitude: 30°). Similar profiles were found throughout the study, with slightly smaller irradiances under fully overcast days (see Figure 3.3).

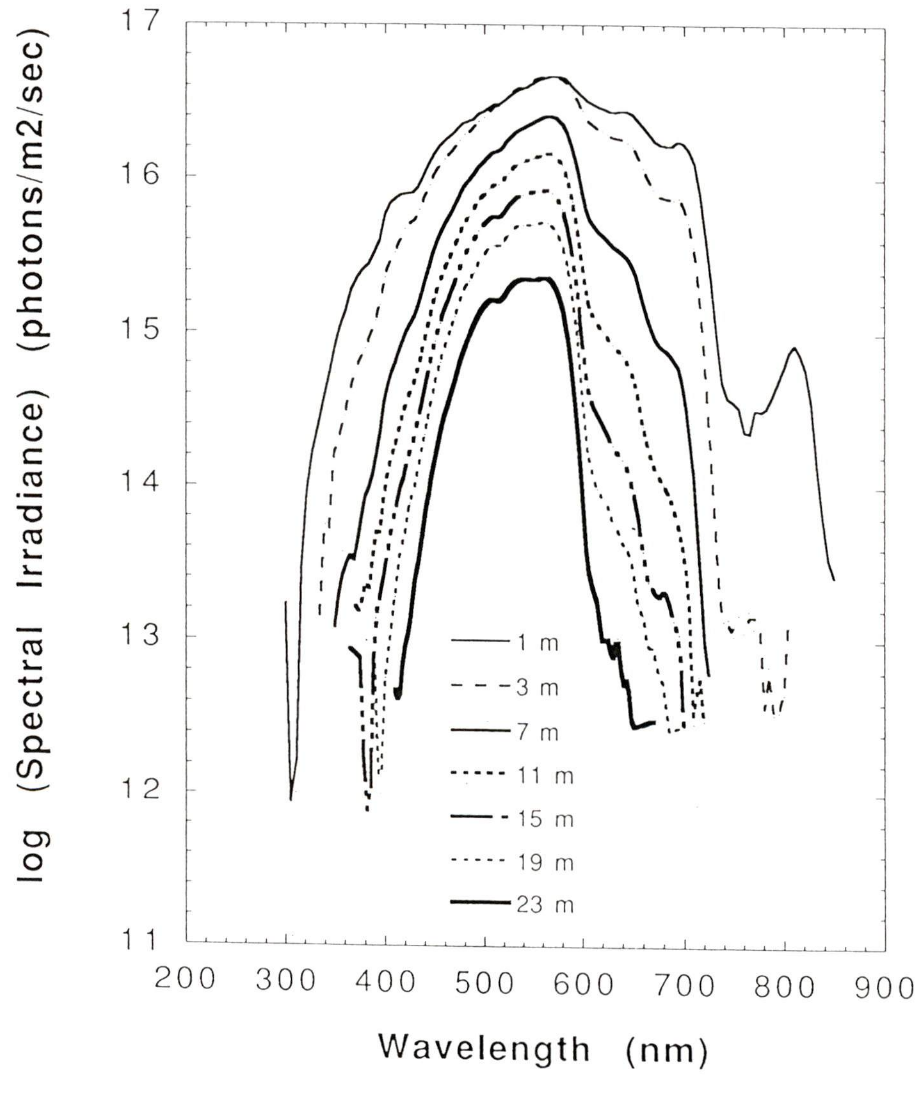


Figure 3.3. Log$_{10}$ of the available spectral irradiance with depth and direction for salmonid vision in Saanich Inlet under clear (n=3) and overcast skies (n=4). Bars on symbols are standard errors of the means. Direction symbols denote the following: UP: upwelling light, HA: horizontal antisun, HS: horizontal sun, and DO: downwelling light.

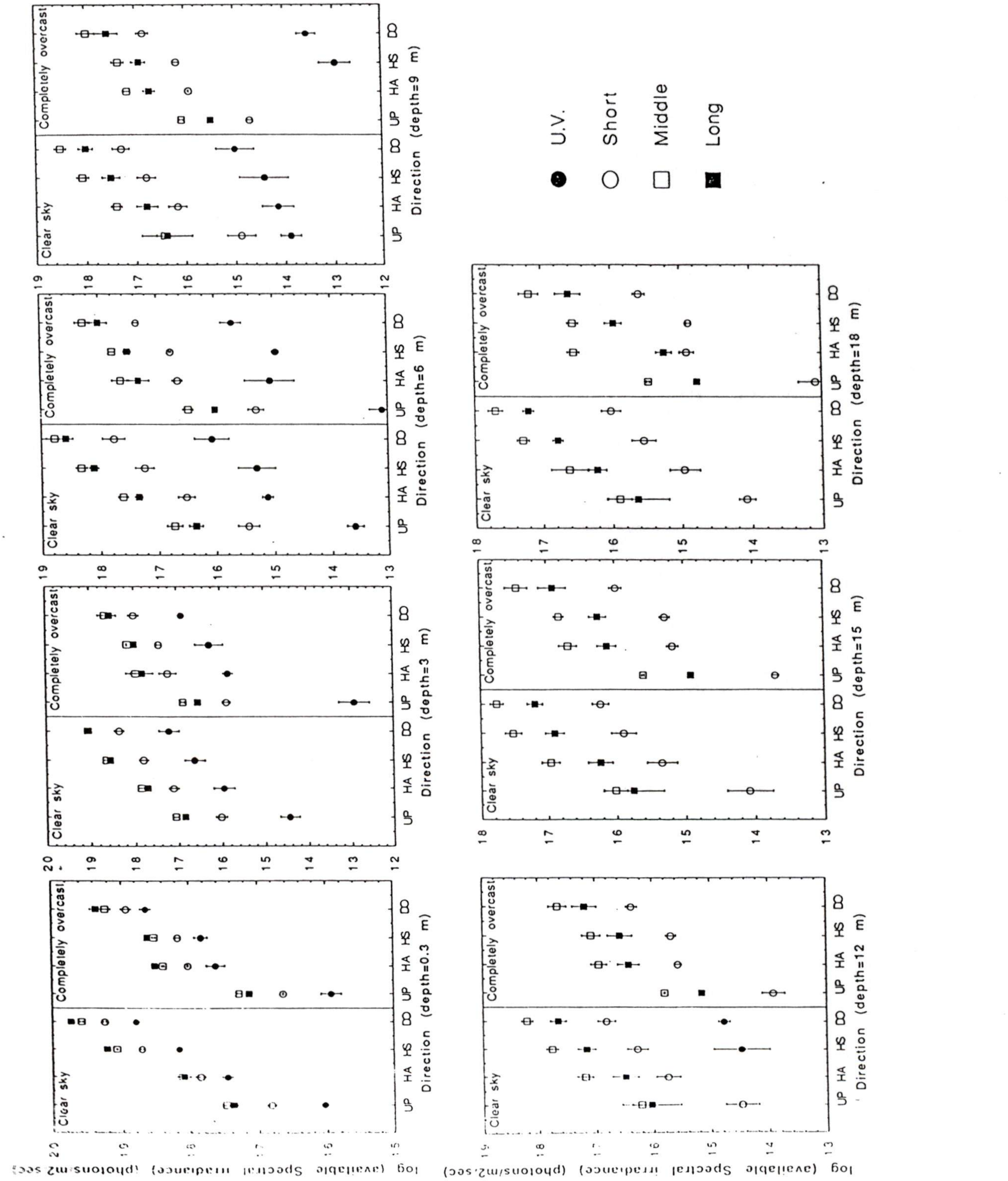


Figure 3.4. Spectral irradiance profiles at 3 meters depth in Deep Cove for various orientations. The scans were taken at 13:00 hours (Pacific Standard Time) on the 16th of July 1991 under clear skies (sun's elevation: 60^{o}).

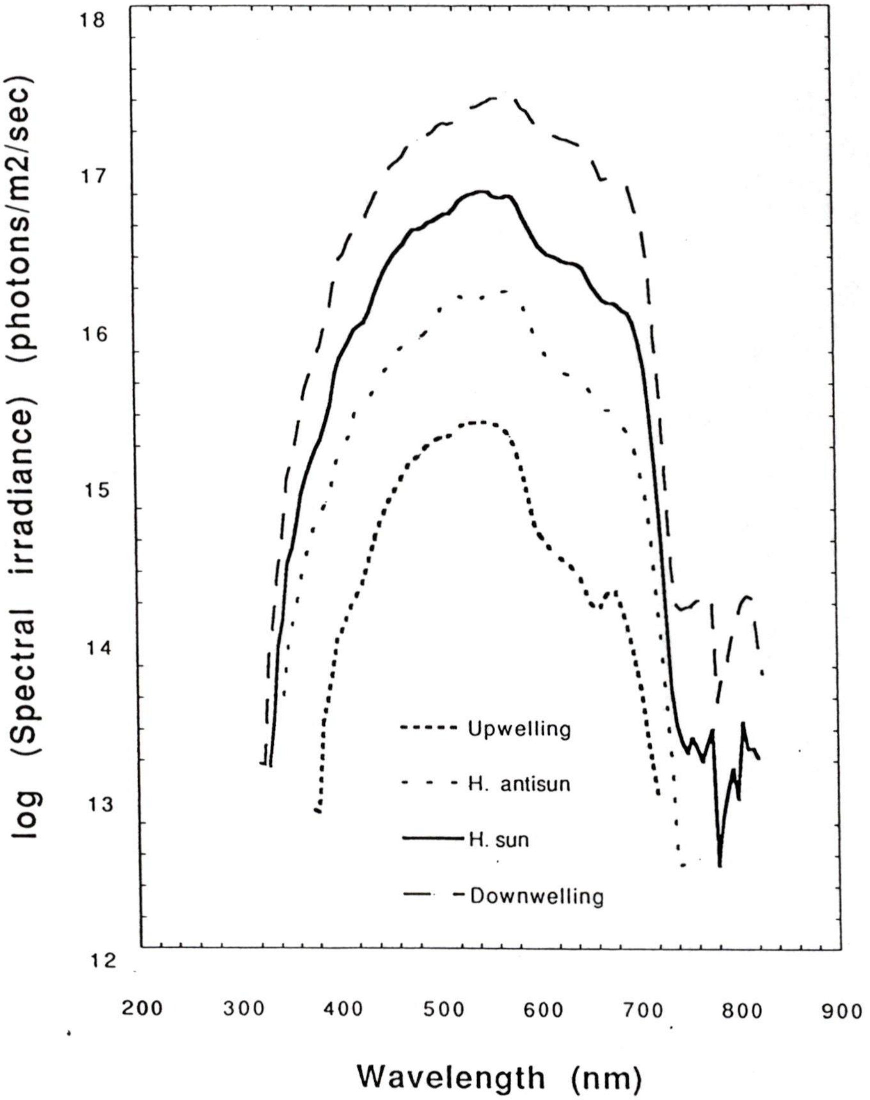


Figure 3.5. Spectral irradiance profiles for Mill Bay taken on the 17th of July 1991 under completely overcast conditions (start of dive: 12:19 hrs, end: 12:44 hrs, Pacific Standard Time). The directionality of the sun's rays was kept through the cloud layer and detected up to 12 meters depth. Decreasing order of light levels as in Figure 3.4.

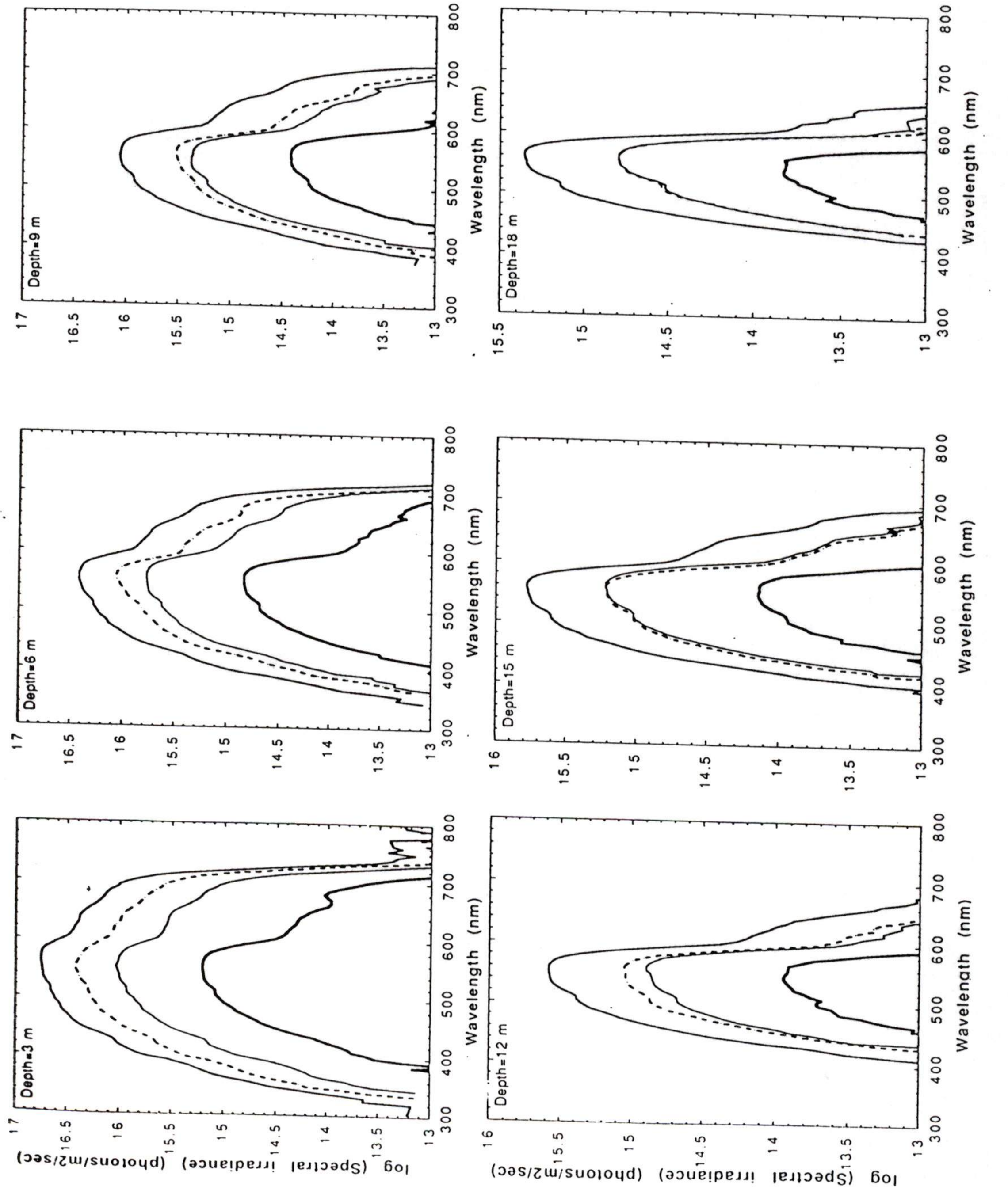


Figure 3.6. Log$_{10}$ of the spectral irradiance corrected for ocular media and ß band absorption of the middle and long wavelength cone mechanisms of small rainbow trout. Direction abbreviations as in Figure 3.3.

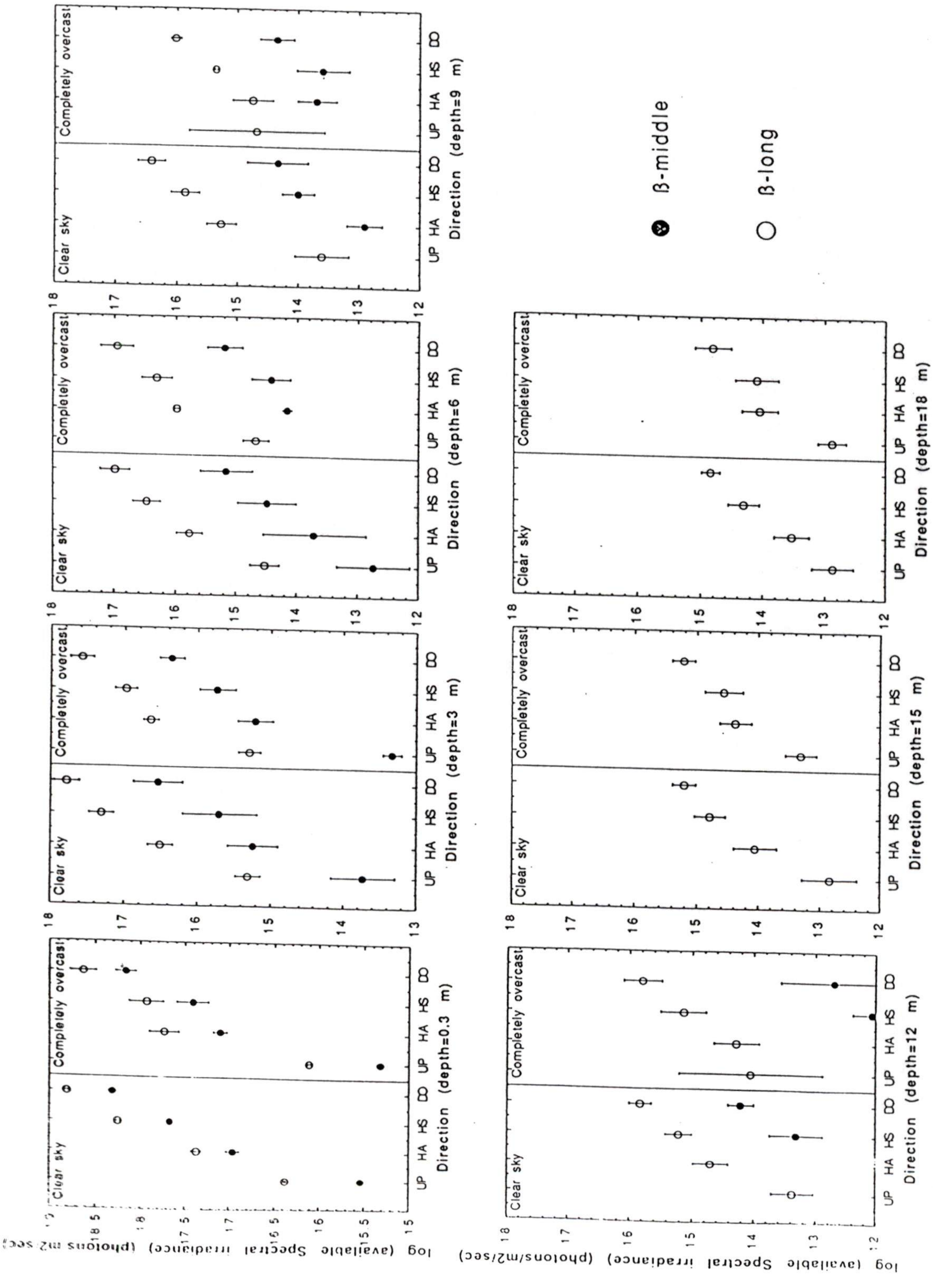


Figure 3.7. Comparison of downwelling available spectral irradiance for salmonid vision in Lake Cowichan, Saanich Inlet and Trevor Channel. Note the higher UV light levels in surface waters of Lake Cowichan. Although more light may penetrate Lake Cowichan, the relative proportions of each type of light remain approximately the same for the three study regions, suggesting similarities in water hue. Particular stations in Saanich Inlet may appear red in colour at times during the summer due to red algal blooms.

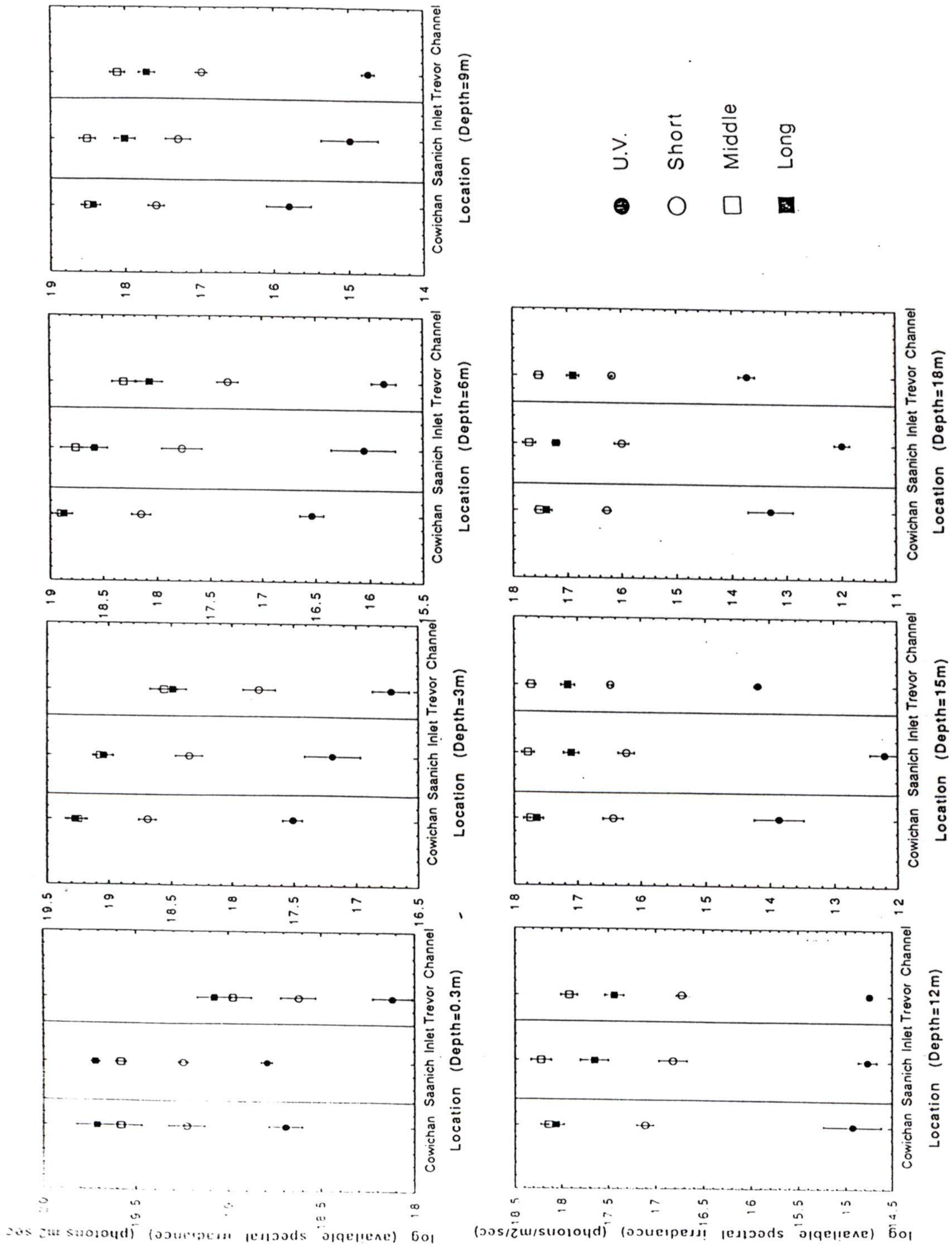


Figure 3.8 (a) Chla concentrations with depth in Lake Cowichan (n=2), Saanich Inlet (n=2) and Trevor Channel (n=1). Trevor Channel and Saanich Inlet have higher concentrations of surface Chla than Lake Cowichan. (b) Number of particles between 0.5 and 12.7 mm in radius in Lake Cowichan and Saanich Inlet. (c) Comparison of DOM levels (presented as Optical Density (OD) data at 350 nm) between Saanich Inlet and Trevor Channel. Chla concentrations for Trevor Channel and all OD measurements were taken from Brown and Borstad (1981). Their study sites V1 and E9 correspond to Saanich Inlet and Trevor Channel, and their measurements were taken at similar times to ours (July-August). Vertical bars depict the standard errors of the means.

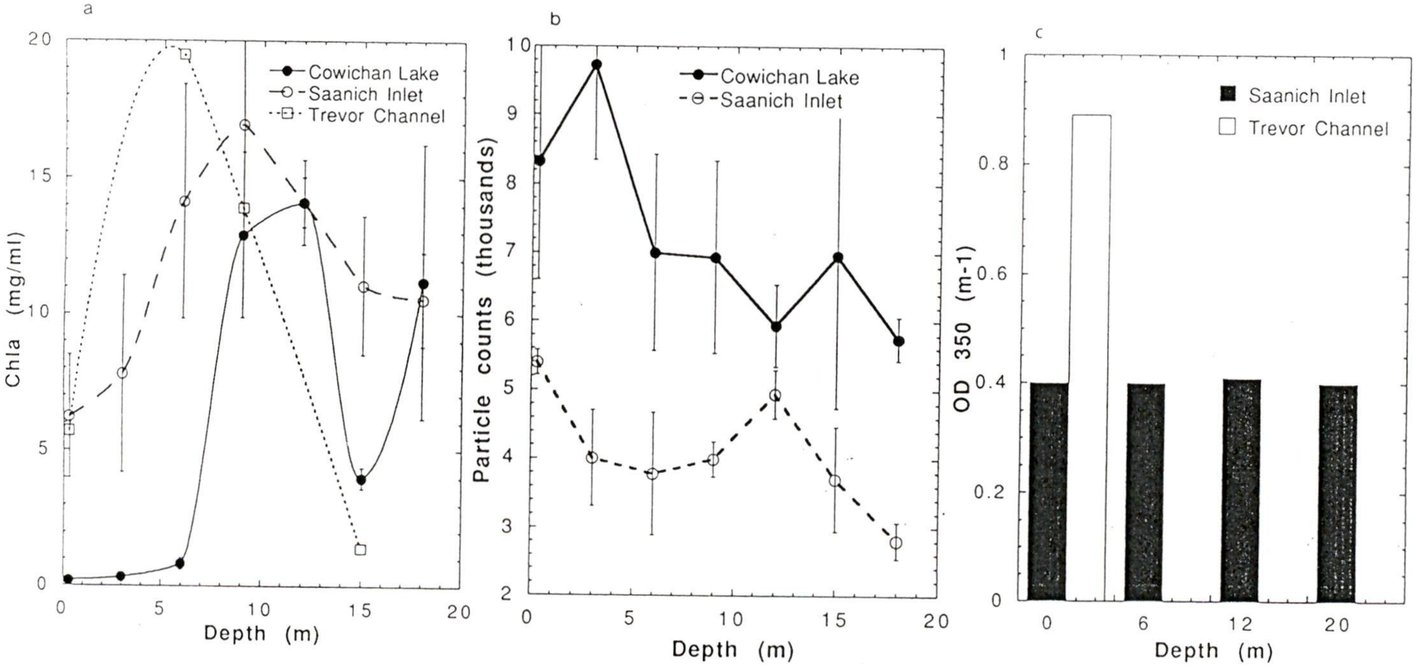
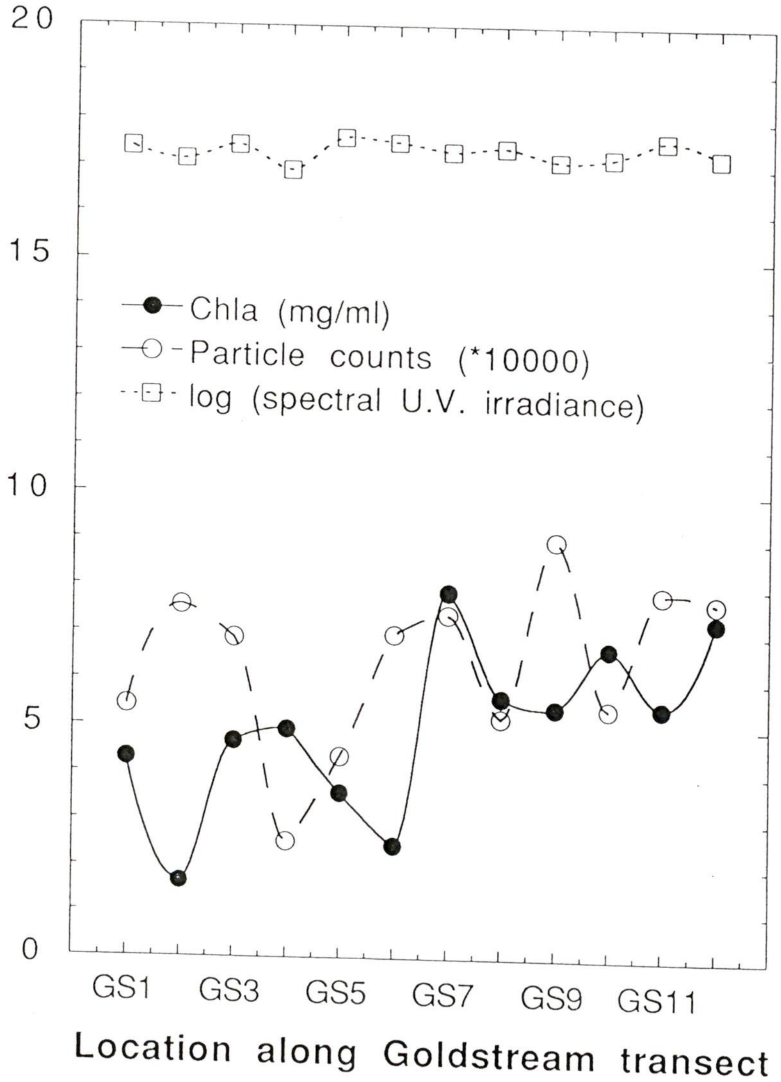


Figure 3.9. Chla concentrations (n=2), particle counts and UV levels along the Goldstream transect. Maxima in Chla concentrations appear to match minima in particle counts generally. Standard errors are within the margin of the data points.



Discussion.

Spectral measurements

The findings presented show that salmonids are restricted to surface waters for visual processes involving UV light (< 15 m in Saanich Inlet). If target contrast is improved with UV vision as suggested by Bowmaker and Kunz (1987), this depth restriction would suggest another reason for the periodic movements of salmon towards the surface to capture prey. Also, salmonids may be approaching the surface to obtain polarized light cues for orientation as mentioned in chapter 1.

Navigation using polarized light has been previously associated with perception of scattered light from patches of sky devoid of clouds (Hawryshyn, 1992). However, although the sun's rays may not reach the ocean directly under a fully overcast sky, navigation using polarized light may still be possible if the directionality of the incoming solar radiation is maintained through the atmosphere (Waterman, 1956). This is because underwater polarization arises mostly from particle scattering in the water column according to the general Mie theory (Van de Hulst, 1957), not from sky polarization (Waterman, 1954, 1955). The most pronounced band of polarized light, produced by "Rayleigh-type" particles would occur theoretically at 90° to the line traced by the incoming radiation passing through the observer (Rayleigh, 1889). Thus, if fish can discern this pattern of polarized light, they could detect the approximate position of the sun behind the cloud layer. The results presented for Saanich Inlet show that directionality of sunlight is maintained under some types of clouds (Fig. 3.5, possibly water droplet clouds, Wellington, 1955) and suggest that polarized light navigation as described above should be possible provided enough polarized UV light for use by fish (salmonids) is present.

The similarities in the shape of the spectral profiles and available spectral irradiances for both coastal waters and Lake Cowichan (Fig. 3.7) suggest that salmonids should not have to change their retinal pigment ratios to maximize their total photon absorbance from one environment to the other (photoperiods and temperatures being approximately equal in both environments during the fish's migration). The preponderance of middle and long wavelengths in both environments would favour a higher porphyropsin/rhodopsin retinal ratio since this would increase absorption of longer wavelengths (Wald, 1939; Bridges, 1972). A change from a rhodopsin- (peak absorption at 503 nm) to a porphyropsin-dominated retina (peak absorption at 527 nm) has been observed for returning salmon of some species prior to entering fresh water (Beatty, 1966). The pigment adaptation to fresh water has been said to be an "anticipation" by the fish; the light measurements presented show that such an anticipation is possibly the result of the spectral similarities between coastal waters and Lake Cowichan.

Nevertheless, some species such as sockeye salmon, O. nerka, (and the landlocked sibling species known as kokanee) always have a rhodopsin-dominated retina at all stages in their life (Beatty, 1966). This situation may reflect the common occurrence of this species of salmon close to the surface, i.e. in a short wavelength rich environment, throughout the life of its individuals (Pella, 1968, French et al., 1976). Rhodopsin based pigments may help sockeye salmon detect prey more easily in the open ocean by acting as offset pigments (contrast hypothesis, Lythgoe, 1968). This situation occurs when the retinal pigment proteins absorb maximally at a wavelength that is not the peak wavelength reflected by the water background and the spectral characteristics of the water background differ with direction of observation (e.g. downwelling vs. horizontally scattered light, see MacFarland and Munz, 1975a). In this case the contrast of a reflective object on the water background (usually composed of longer wavelengths) would be more readily detected than by having a pigment whose maximum absorption matches the background (this because the offset pigment will be less sensitive to the background illumination, McFarland and Munz, 1975a). In general, offset pigments reduce photosensitivity to available light but increase contrast of reflective targets (Lythgoe, 1968); matching pigments, on the other hand, increase photosensitivity to available light and are therefore good shadow target detectors. In coastal waters, a rhodopsin dominated retina would only increase the target detection capabilities of sockeye salmon if the reflecting target, viewed in the horizontal direction, preferentially scatters shorter wavelengths (since downwelling and horizontal irradiance have the same overall spectral composition, Fig. 3.4). This may be the case for schools of small herring which "flicker" as they change orientation in surface waters (these schools being a major source of food for returning salmon, Wing, 1977, Healey, 1980). The approximate 0.7 log difference between downwelling and horizontal antisun light at 503 and 527 nm (Fig. 3.4) could also be used by fish with rhodopsin or porphyropsin dominated retinae to detect targets on the basis of intensity (the target reflecting downwelling irradiance over a dimmer horizontal background). To this regard, a porphyropsin dominated retina may act as offset or matching the background illumination depending on the spectral characteristics of the water body. However, the wavelength of maximum absorption of its proteins being higher than those forming the rhodopsin dominated retina, its protein-based pigments would be less offset when operating in coastal waters than those of a rhodopsin dominated retina (Fig. 3.4, maximum transmission at 577 nm).

Water chemistry

The available spectral irradiances found in coastal waters and Lake Cowichan are in good agreement with what would be expected from their biophysical properties. UV light is most abundant in Lake Cowichan as the Chla concentrations are the lowest at this location (Figs. 3.7 and 3.8a). Within coastal waters, Trevor Channel has the highest concentrations of DOM and Chla near the surface, which explains the lower UV levels as compared to Saanich Inlet (Figs. 3.7 and 3.8a,c). However, Saanich Inlet stations had higher average concentrations of Chla than Trevor Channel for depths greater than 10 meters. This may explain the accelerated decrease in UV light for Saanich Inlet at such depths (Fig. 3.7).

The relative numbers of particles and concentrations of Chla along the Goldstream transect (Fig. 3.9) may be due to a layer of freshwater resting over the more saline coastal water. Two days of stormy weather preceded these measurements and brownish coloured water from runoff was often detected near the shore-line (especially as river outflows were approached). Collection of surface samples would thus contain large numbers of particles which may not yet have mixed and reached an equilibrium with the marine phytoplankton. The small particle sizes counted further indicated these to be runoff nutrients which could be consumed by moderate size phytoplankton. This might explain the opposite fluctuations found between particle numbers and Chla concentrations along the Goldstream transect.

UV intensities at the various transect stations tended to decrease with increasing Chla concentrations, but the negative regression between these two variables for the two transects was not significant (F-test probability=0.6). Similar results have been reported by Gower and Borstard (1990) for Barkley Sound (Vancouver Island). These authors suggested the presence of large and variable amounts of DOM of terrigenous origin to explain the lack of correlation; the results found and observations made during this study also support this hypothesis. Chla concentrations, like DOM levels along Vancouver Island's coast, are variable and dependent on many factors such as currents, upwellings, river inputs and types of catchment area (Mackas, 1992; Pan et al., 1988). It is therefore unlikely that, once having entered coastal waters, the fish could follow any trends in UV light levels to reach the spawning rivers. This suggests that the fish would be using polarized light patterns (not differences in irradiances) for orientation if using photic cues during migration. Since Chla concentrations during the early spring and late fall are, on average, lower than those found in this study (Chla concentrations in August are seven times those in March or April for the Inner Shelf, comparable for Juan de Fuca Strait and at least five times those in October or November for both regions, Mackas, 1992; Pan et al., 1988), sufficient light for stimulation of UV photoreceptors should also be available during peak migratory periods. To our knowledge, there is no seasonal information on DOM levels for Vancouver Island waters; but, Mackas' (1992) results for dissolved nitrate show its mean concentrations to be lower during spring and fall than during the summer for Juan de Fuca Strait, and lower during the spring but higher during the fall for the Inner Shelf (note, however, the variations during the summer for this region).

It is interesting to compare the irradiance values found in coastal waters with those representative of clear open ocean waters (Antartic Ocean, Prezelin et al., 1991) and those predicted by models for waters with different concentrations of Chla (Baker and Smith, 1982). The diffuse attenuation coefficient calculated for waters with low Chla content (<0.5 mg/ml) in the region from 350 to 400 nm is three to twenty times lower than that of the coastal waters studied (Baker and Smith, 1982). Similarly, extrapolated and measured values in this region of the spectrum for clear ocean waters show irradiances that are 100 to 1000 times higher than those reported in this study (Baker and Smith, 1982, Prezelin et al., 1991). Assuming an attenuation coefficient of 0.1 m^{-1} (representative of clear waters with Chla concentrations of 0.05 mg/ml) the depth for UV cone stimulation would be around 96 meters. If the attenuation coefficient is lowered to 0.07 m-1, this depth becomes 137 meters. Clearly, there is plenty of UV light for salmonid visually-mediated behaviour in the open ocean.

In summary, the measurements obtained show that there is sufficient UV light for visually-mediated salmonid behaviour to around 15 meters in depth in coastal waters of Vancouver Island. UV light levels do not follow trends with distance from the spawning rivers but appear to be related to phytoplankton concentrations and DOM levels, themselves variable throughout the coast-line. The results also show that directionality of the incoming light is maintained under certain types of cloud suggesting that polarized light navigation may still be possible under overcast skies provided sufficient UV polarized light is present in surface waters.

Chapter 4

Use of Mie theory to approximate the radiance distribution in a lake (a simplified approach)

Introduction

Most studies describing the angular distribution of scattered radiation in natural water bodies are based on measurements of the volume scattering function (Sasaki et al., 1962, Duntley, 1963, Ochakovsky, 1966, Kirk, 1991). This function defines the proportion of incident radiation scattered within a volume determined by the intersection of the beams of incident and scattered light (Spielhaus, 1965). The volume scattering function has been widely used as a characteristic parameter to classify water bodies (Jerlov, 1976 pp. 36-37).

Attempts at explaining the behaviour of light traversing a medium containing scatterers have been put forward for artificially-created suspensions of different size particles in laboratory conditions (Heller and Tabibian, 1962, Hepplestone and Lewis, 1968). Mie's scattering theory applied to such systems has resulted in fairly good agreement between computed and measured scattered light intensities (Hepplestone and Lewis, 1968); however, in no case do these laboratory experiments approximate the bio-chemical complexity of natural water bodies. It is perhaps due to this complexity that very few authors have tried to apply scattering theory to explain their results obtained in natural waters (Morel, 1973). Furthermore, in such cases, disturbance to the water samples due to necessary handling (e.g. pumping ocean water into a research vessel) may have altered the optical properties of the water and therefore biased the results. Only insitu underwater measurements can overcome these difficulties.

In this study, the LI-1800 underwater spectroradiometer was used to measure the radiant energy scattered at two angles (49° and 131°) to the refracted incident sunlight impinging on Lake Cowichan. Radiance measurements and water samples were taken by SCUBA divers at three meter intervals from 9 to 0.3 meters from the surface. The water samples were processed using a Coulter counter to determine the size and concentration of particles with defined radii from 0.39 to 3.17 μm. Knowing the direction and intensity of the downwelling radiant energy and assuming an average relative refractive index for the set of particles counted, Mie theory was applied to approximate the scattered light intensities at both angles. The calculations neglect molecular scattering and do not partition the radiance reaching the cosine collector into its various components (direct vs. diffuse radiance). Furthermore, the approach is not based on calculation of the volume scattering function, nor does it follow conventional procedures based on radiative transfer theory. Instead, the predictions are based solely on scattering by particles within a specific distance from the spectroradiometer cosine collector, determined by its sensitivity.

Materials and Methods

Spectral radiance measurements

In an attempt to restrict the light gathered to a specific angle, a cone 13.5 cm in length and sustaining an angle of 30° at the apex was installed over the spectroradiometer cosine collector. The cone's interior was painted black to maximize absorption of stray light that could have otherwise reached the cosine collector by multiple internal reflection off the cone's sides. Upwelling light from reflection off the lake's floor was insignificant because the bottom of the lake was deeper than 20 meters.

SCUBA divers took spectral radiance measurements from 9 to 0.3 meters depth at three meter intervals in lake Cowichan during late February and early March 1992. Immediately before diving, the sun's altitude was measured with a sextant (30° in both dives), and Snell's law was used to correct for refraction of the sun's rays at the air-water interface. Three scans ranging from 300 to 850 nm were taken at each depth. The scans show spectral radiance of incident downwelling light, and light scattered at 49° and 131° to it ((spectroradiometer pointing horizontally in the water column towards the sun and away from it (antisun direction) respectively), Fig. 4.1). The procedure for data collection has been outlined in chapter 1.

Particle counts and Chlorophyll a measurements

Estimates of the relative amounts of particulate matter at each depth were obtained using a Coulter Counter (model TA II, Coulter Electronics Inc., Hialeah, Florida). Using a 70 µm aperture tube, particles with the following radii were counted in 0.05 ml samples: 0.39, 0.5, 0.63, 0.79, 1, 1.26, 1.59, 2, 2.52 and 3.17µm . Particles in this size range (1-2 µm) produce most of the scattering in the Sargasso and Mediterranean seas (Kullenberg, 1968, 1974), results which have been supported by laboratory findings (Zaneveld et al., 1974).

Water samples form each depth were processed for concentrations of Chla using the procedure in Jeffrey and Humphrey (1975).

Calculation procedure

The amount of scattered light in the direction ø from a randomly polarized beam of unit intensity can be calculated using the following equation (Mie, 1908):

$$I(ø)=\partial^2(i_1+i_2)/(8\pi^2)$$

where ∂ is the wavelength of the incident light and, i_1 and i_2 (the scattered components with Electric vectors perpendicular and parallel to the plane of observation[1]) are given by:

$$i_1=[\Sigma\{A_n\pi_n+P_n[\Omega\pi_n-(1-\Omega^2)\pi'_n]\}]^2$$

$$i_2=[\Sigma\{A_n[\Omega\pi_n-(1-\Omega^2)\pi'_n]+P_n\pi_n\}]^2$$

In these formulae:

$\Omega=\cos ø$, where ø is the angle between the direction of propagation of the scattered light and the reversed direction of propagation of the incident light,

π_n and π'_n are the Legendre polynomials and their derivatives of order n. These can be calculated from the recurrence formulae:

$$\pi_n=\Omega[(2n-1)/(n-1)]\pi_{n-1}-[n/(n-1)]\pi_{n-2}$$

$$\pi'_n=(2n-1)\pi_{n-1}+\pi'_{n-2}$$

with $\pi_0=\pi'_0=\pi'_1=0$.

---

[1] The plane of observation is defined as the plane containing the direction of observation and the direction of propagation of the incident light.

The An's and Pn's are functions involving Riccati-Bessel and Riccati-Hankel functions, and are related to the Bessel functions of half integral order:

$A_n=[(-1)^{n+1/2}(2n+1)/(n(n+1))][a_n]$

$P_n=[(-1)^{n+3/2}(2n+1)/(n(n+1))][b_n]$,

where,

$a_n=[(\pounds\yen'_n(ß)\yen_n(\pounds)-ß\yen'_n(\pounds)\yen_n(ß))/(\pounds\yen'_n(ß)f_n(\pounds)-ßf'_n(\pounds)\yen_n(ß))]$

$b_n=[(ß\yen'_n(ß)\yen_n(\pounds)-\pounds\yen'_n(\pounds)\yen_n(ß))/(ß\yen'_n(ß)f_n(\pounds)-\pounds f'_n(\pounds)\yen_n(ß))]$

$ß=m\pounds$, with $\pounds=2\pi r/\partial$, r being the radius of the particle and m the index of refraction of the particle relative to that of the surrounding medium.

The major computational problem relies in evaluating a_n and b_n. However, defining $A_n(ß)=\yen'_n(ß)/\yen_n(ß)$, these terms can be re-written (Wickramasinghe, 1973):

$a_n=\{[A_n(ß)/m+n/\pounds]\,\mathrm{Re}\{f_n(\pounds)\}-\mathrm{Re}\{f_{n-1}(\pounds)\}\}/[(A_n(ß)/m+n/\pounds)f_n(\pounds)-f_{n-1}(\pounds)]$

$b_n=\{(mA_n(ß)+n/\pounds)\,\mathrm{Re}\{f_n(\pounds)\}-\mathrm{Re}\{f_{n-1}(\pounds)\}\}/[(mA_n(ß)+n/\pounds)f_n(\pounds)-f_{n-1}(\pounds)]$,

and the $f_n(\pounds)$ and $A_n(ß)$ can be calculated using the following relations:

$f_n(\pounds)=((2n-1)/\pounds)f_{n-1}(\pounds)-f_{n-2}(\pounds)$

$A_n(ß)=-n/ß+(n/ß-A_{n-1}(ß))^{-1}$

with $f_0(\pounds)=\sin\pounds+i\cos\pounds$, $f_{-1}(f)=\cos\pounds-i\sin\pounds$, and $A_0(ß)=\cos ß/\sin ß$.

The previous equations were used to calculate i_1s and i_2s for each particle radius measured with the Coulter counter. Convergence of the sums was fairly rapid because of the small £ values. Where applicable, our results were confirmed against tables of light scattering functions for spherical particles previously published by the National Bureau of Standards (1949), the University of Michigan Engineering Research Institute (1951) and Wickramasinghe (1973).

In order to calculate the total scattered intensity at angle ø, the contribution from all the particles that scattered within the 30° cone defined by the radiance collector was summed (the cone surrounding the cosine collector surface held an apex angle of 30°, see Figure 4.2). The distance within which a particle had to scatter in order for the light to be detected by the cosine collector was limited by the sensitivity of the instrument (10^{-5} µmoles/m^2/sec). This threshold distance was calculated knowing the attenuation coefficient of the lake water at each wavelength using the formula (Tyler and Preisendorfer, 1962)

$$d=-(1/k)\log(I_2/I_1),$$

where I_1 and I_2 are incident and outgoing light intensities traversing a distance d of water. The attenuation coefficients for each wavelength were generated using the same formula from downwelling radiance measurements at 0.3 and 9 meters (Table 4.1). The threshold distances are the limits from which a beam of unit intensity would contribute a measured radiance of 10^{-5} µmoles/(m^2*sec*sr).

Knowing the previous parameters, the contribution to the total scattered intensity at angle ø by each particle size (I_p) was deduced from

$$Ip = (I)(I(ø))(c)\int \exp(-kd)dV,$$

where c is the concentration of particles of radius r, I is the incident radiance of wavelength ∂, I(ø) is the scattered intensity per unit incidence (described earlier), and the integration is throughout the volume of the imaginary cone with length d (Fig. 4.2).

The previous method for calculation of the total scattered intensity at angle ø can be used, provided a correction is made for the differential change in particle numbers with increasing volume. The threshold distance for particle scatter was based on the assumption that the light scatter came from particles that were placed along a line normal to the cosine collector (Fig. 4.2). Thus, for every infinitesimal element of length dl, the particles contained within the volume limited by the cone radius were assumed to scatter as if along a line normal to the cosine collector. This is obviously wrong because the integrated volume is not a cylinder. With increasing threshold distance, the extra particles in the integrated volume will contribute an error proportional to the ratio of the square of their radii (Fig. 4.2). Also the total error between two threshold distances with radii r_1 and r_2 will be given by $\int(k_2/k_1)^2 dl$, where the limits of the integral are 10^{-5} and 1 μmole/(m^2*sec*sr) (unit light intensity being detected by the instrument). The light scattered at each wavelength was corrected with respect to 360 nm, by multiplying the scattered intensities by the factor $(k_{\partial}/k_{360})^2$.

The above analysis was performed for the following wavelengths: 360, 380, 400, 440, 480, 520, 560, 600, 640 and 680 nm.

Assumptions

Mie's formulae apply to scattering by spherical particles. It is obvious that this is an ideal case that does not hold for all particles in natural waters; however, in the case of irregular non-absorbing randomly oriented particles, the scattering can be attributed to that produced by spherical particles with the same projected area (Jerlov, 1976 pp. 28). It was therefore assumed that the particles counted were inorganic (which absorb light minimally). This assumption is consistent with the low phytoplankton (Chla) levels found in Lake Cowichan during the study (Table 4.2, although these are much higher than in the open ocean). Phytoplankton cells and their decay products constitute the organic component of the lake's water responsible for light absorption; their size range is usually bigger (5-70 µm range) than the size of the particles counted. A relative refractive index of 1.2 was assumed for the ensemble of particles; this value is within the range described by researchers studying inorganic matter (Pavlov and Grechushnikov, 1966).

The application of Mie's theory to scattering by a system of particles further requires that these be independent, i.e. that the distance between neighbour particles be at least three times their radii (Van de Hulst, 1957). In this case, the intensities scattered by each particle can be added to give the total scattered intensity. Judging from particle counts found with the Coulter counter, this assumption appears to hold for the waters of Lake Cowichan (Table 4.3).

In calculating the threshold distance within which photons must be scattered in order to be detected by the spectroradiometer, vertical extinction coefficients were used for light travelling horizontally or in oblique directions (scattered light). This simplification may be used provided the waters are approximately homogeneous with depth. The chlorophyll concentrations in Table 4.2 support this assumption; furthermore, this study took place in epilimnetic waters of the lake, so drastic changes with deeper water domains (metalimneon and hypolimneon) were avoided (Wetzel, 1975).

Directionality of the sun's rays after having penetrated the water was assumed to be maintained for the depths studied. Measurements at both scattering angles show that this is generally the case (Fig. 4.1), although the light rays become more normal (perpendicular) to the water surface with increasing depth. For this reason, the limiting depth for measurements was set at 9 m.

Results

Given the above assumptions, predictions using Mie theory can be seen to differ up to 8.31 fold with measured light intensities (∂=640 nm, Fig. 4.3, 3m depth). However, an average of 3.76 times (0.57 log units) was obtained for the total data set (Figs. 4.3 and 4.4). Standard errors from particle counts varied from 4.1 to 14.5% (Table 4.3) and account for part of the total error in the predicted intensities. Nevertheless, because the remaining errors arising from the change in directionality of the sun's rays with depth, the uncertainty in scattered intensity produced by particles outside the range counted and at neighbouring angles to the one measured, and errors in the predicted A_n and P_n functions from assuming a general refractive index for all the particles are unknown, the overall partition of errors cannot be gauged.

Predicted and observed radiances agree overall better at a scattering angle of 131° (Fig. 4.3-4.5). When the particles doing the majority of the scattering are bigger than 1/10th of the wavelength of the incident light (non "Rayleigh particles"), Mie's theory predicts an increase in the light scattered in the forward direction, thus supporting this result. Scattering at smaller angles (e.g. 49°) is partly accomplished by Rayleigh particles, and, in this case, the scattering is inversely proportional to the 4th power of the wavelength. This would argue for smaller predicted than measured radiances in the UV wavelengths (330-450nm), a situation which is generally not observed (Figs. 4.4 and 4.5). This may indicate that scatter by smaller particles is not predominant in Lake Cowichan, as observed for other water bodies (Zaneveld et al., 1974), or it may be a consequence of the relative refractive index (see discussion).

The great similarity in the results obtained for both months can be observed by comparing the slopes between predicted and observed radiances for each month (Fig. 4.5). This similarity agrees well with analogous findings comparing particle counts and phytoplankton concentrations.

Table 4.1. Attenuation coefficients for each wavelength for both months studied.

∂(nm)	360	380	400	440	480	520	560	600	640	680
k(m^{-1})										
February	1.31	1.16	1.09	0.64	0.49	0.31	0.29	0.35	0.62	0.69
March	1.19	1.03	0.82	0.58	0.49	0.42	0.44	0.45	0.45	0.49

Table 4.2. Chla concentrations and standard errors (in brackets) for both dives. Values obtained from averages of two samples.

Depth (m)	0.3	3	6	9
Chla (mg/ml)				
February	0.61 (0.052)	0.44 (0.12)	1.15 (0.55)	0.12 (0.08)
March	1.4 (0.09)	0.59 (0.35)	1.24 (0.93)	0.69 (0.5)

Table 4.3. Sample particle counts and associated standard errors (in percent of the mean value) for 3 m depth. Values calculated from averages of 5 samples. Standard errors from other depths were smaller or equal to the ones on this table.

r(μm)	0.39	0.5	0.63	0.79	1	1.26	1.59	2	2.52	3.17
Counts										
Feb.	8227	13699	22229	31718	41525	58340	22273	3039	1562	234
S.E.%	13.1	8.6	5.9	14.5	11.3	9.7	11.6	4.3	7.1	6.5
Mar.	5319	11754	72345	49529	40753	66521	40246	6319	1095	435
S.E.%	7.2	12.4	14.1	13.2	8.6	12.3	7.9	5.7	6.4	9.8

Figure 4.1. Spectral radiance with depth in Lake Cowichan during early March 1992. Top, middle and lowest radiance curves correspond to scans taken in the incident light's direction, at 131° and at 49° to it. (see Figure 2.2 in chapter 1) Note the differences in radiance for the 131° and 49° curves, which demonstrates that a certain directionality of the incoming radiation at the surface of the lake is maintained under water. The ripples in the top surface scan were caused by waves.

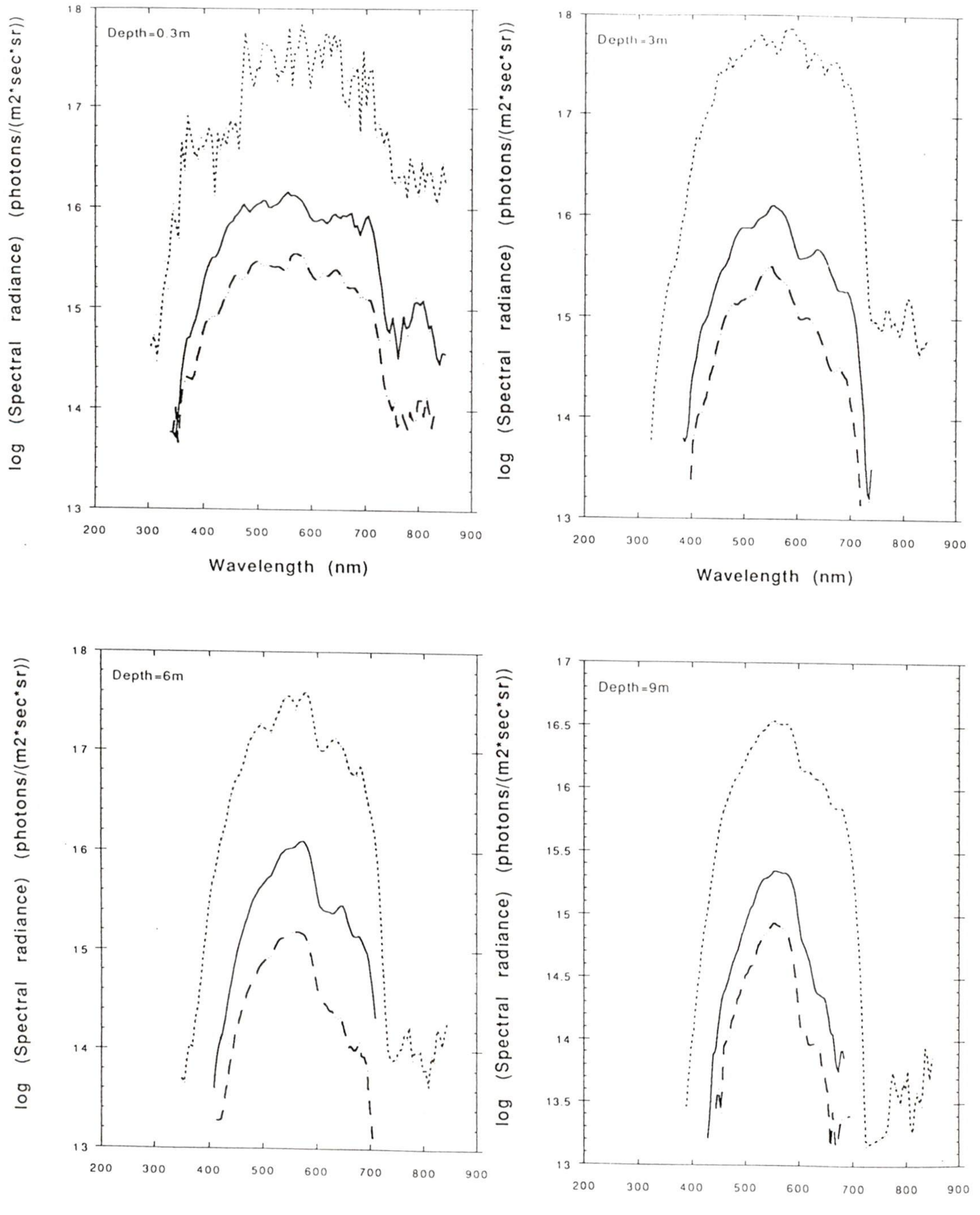


Figure 4.2. A view of the spectroradiometer with its light gathering cosine collector (half moon structure), and the cone that restricted incoming light to an angle of 30°. The picture also shows a hypothetical volume determined by a given threshold distance within which a unit light of a specific wavelength must be scattered in order to reach the cosine collector and be detected (10^{-5} μmoles/(m^{2}*sec*sr)). From the picture, $r_1=d_1*\tan15°$ (the extra distance within the spectroradiometer from the cosine collector to the cone apex is insignificant compared to calculated ds) , and the volume up to this infinitesimal cylinder will be given by: $\int\pi r_1{}^2 dl = \int\pi(d_1\tan15°)^2 dl$. Because the integrated volume is not a cylinder (dashed lines), the method of summing the contributions from each particle within the volume to obtain the total scattered intensity biases the results. With increasing threshold distance d, more "extra particles" (particles outside the dash cylinder) will contribute to the scattered radiance;this will give higher scattered radiances for wavelengths with smaller attenuation coefficients. The ratio of scattered radiances contributed by "extra particles" in infinitessimal toroid volumes with radii r_1-2.5 cm and r_2-2.5 cm is proportional to $\int(d_1/d_2)^2 dl$ (using previous geometry and calculus; 2.5 cm is the diameter of the cosine collector). But d is inversely proportional to k, the extinction coefficient; thus the previous ratio translates to $\int(k_2/k_1)^2 dl$. Because 360 nm gives the smallest d, all other wavelength radiances were corrected with respect to it.

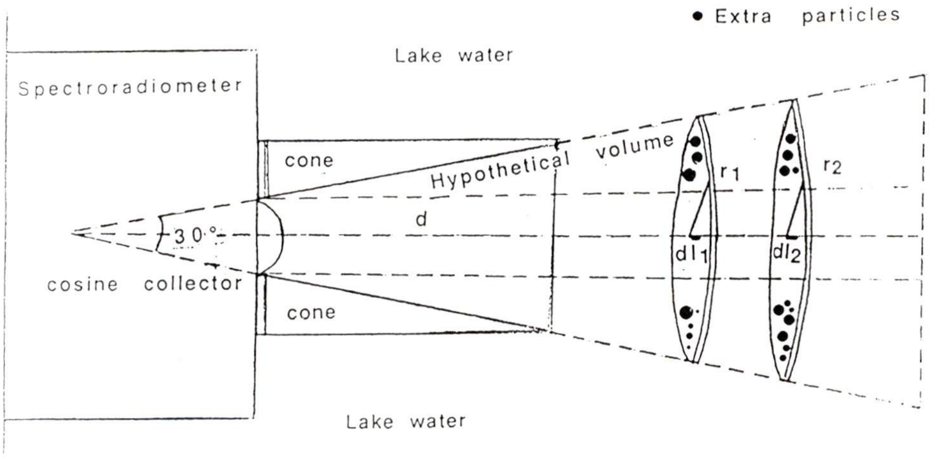


Figure 4.3. Measured and predicted $\log_{10}$ spectral radiances for late February. Dark circles and white circles are predicted and measured radiances at 131°, dark squares and white squares are predicted and measured radiances at 49°.

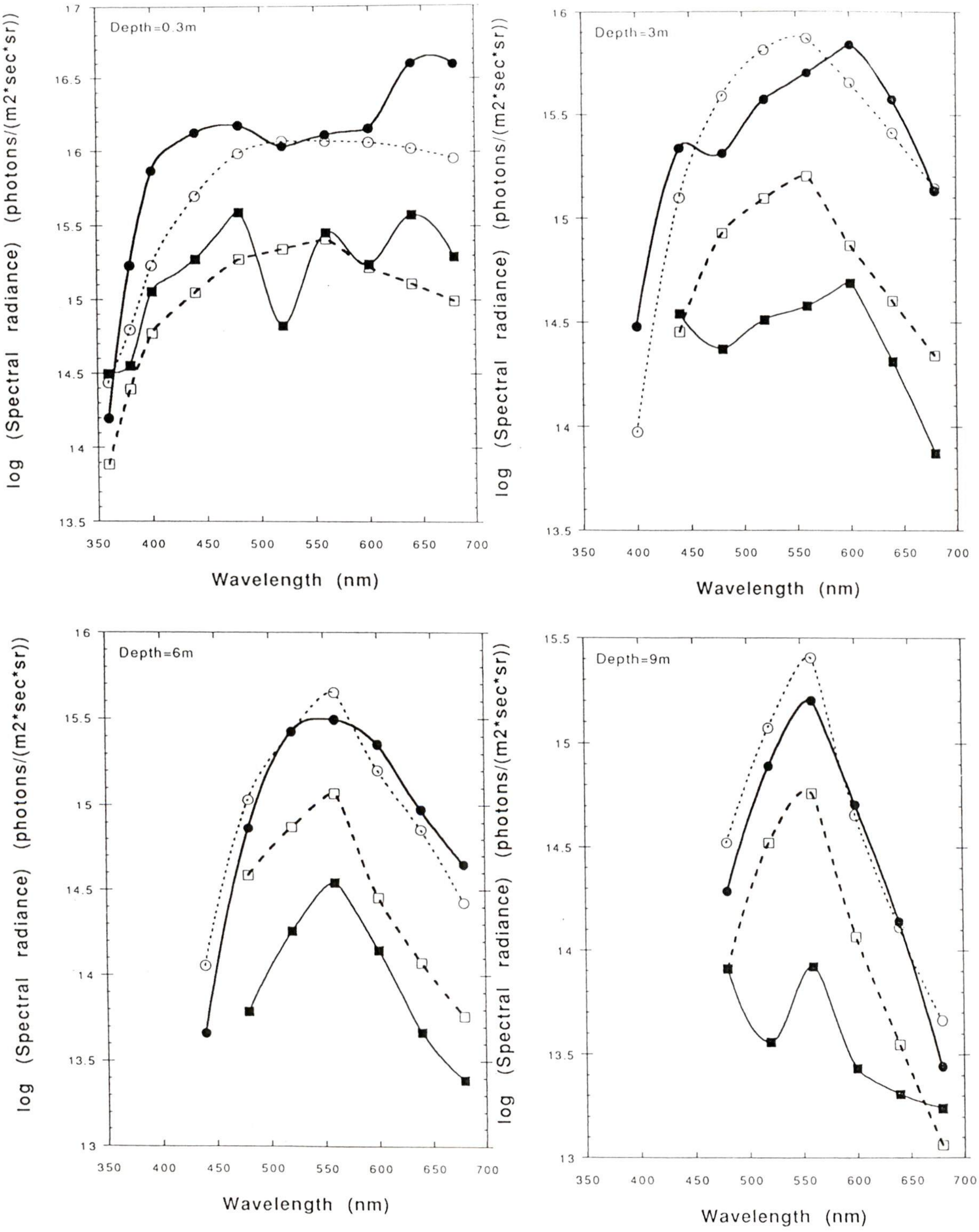


Figure 4.4. Measured and predicted $\log_{10}$ spectral radiances for early March. Same notation as in Figure 4.3.

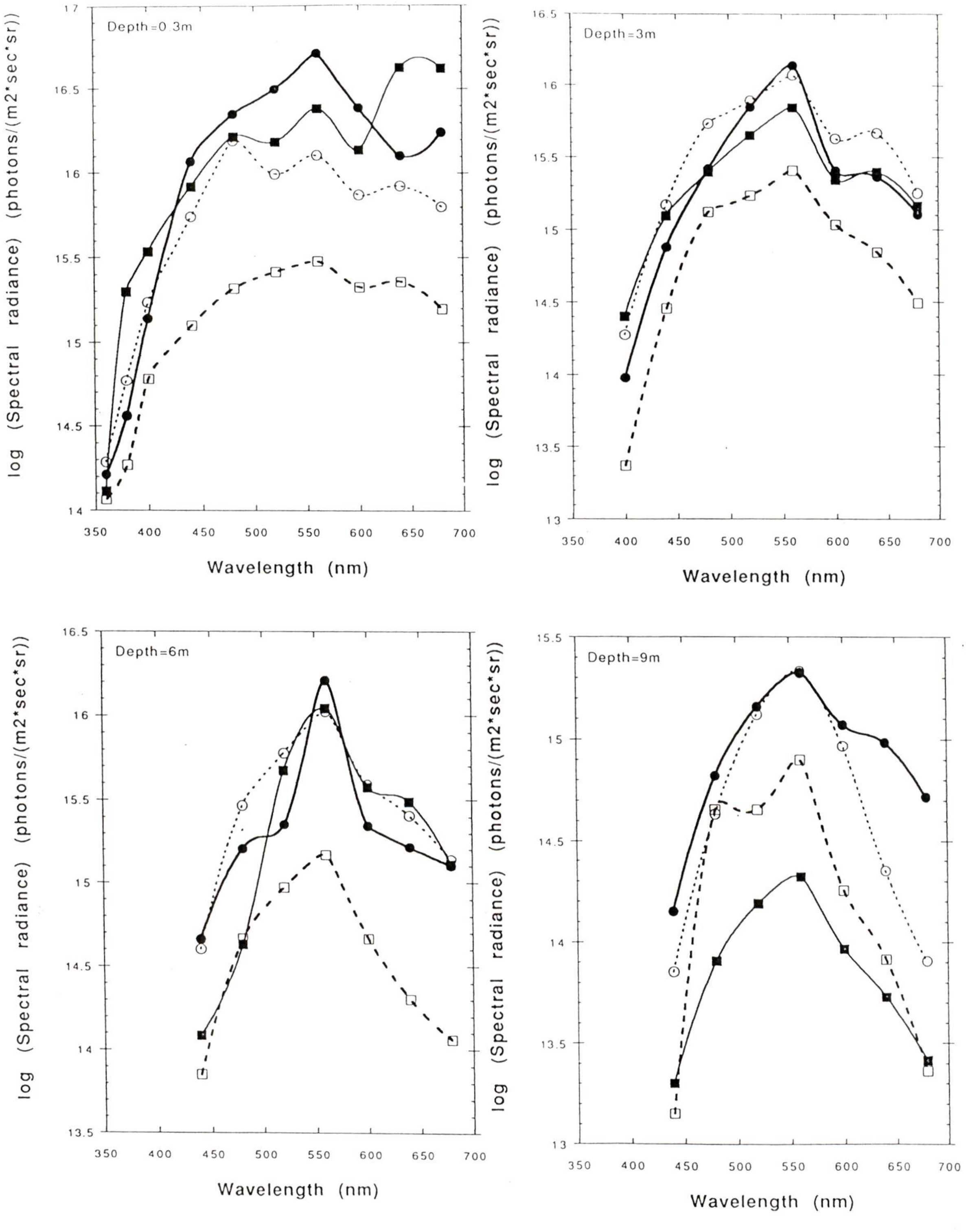
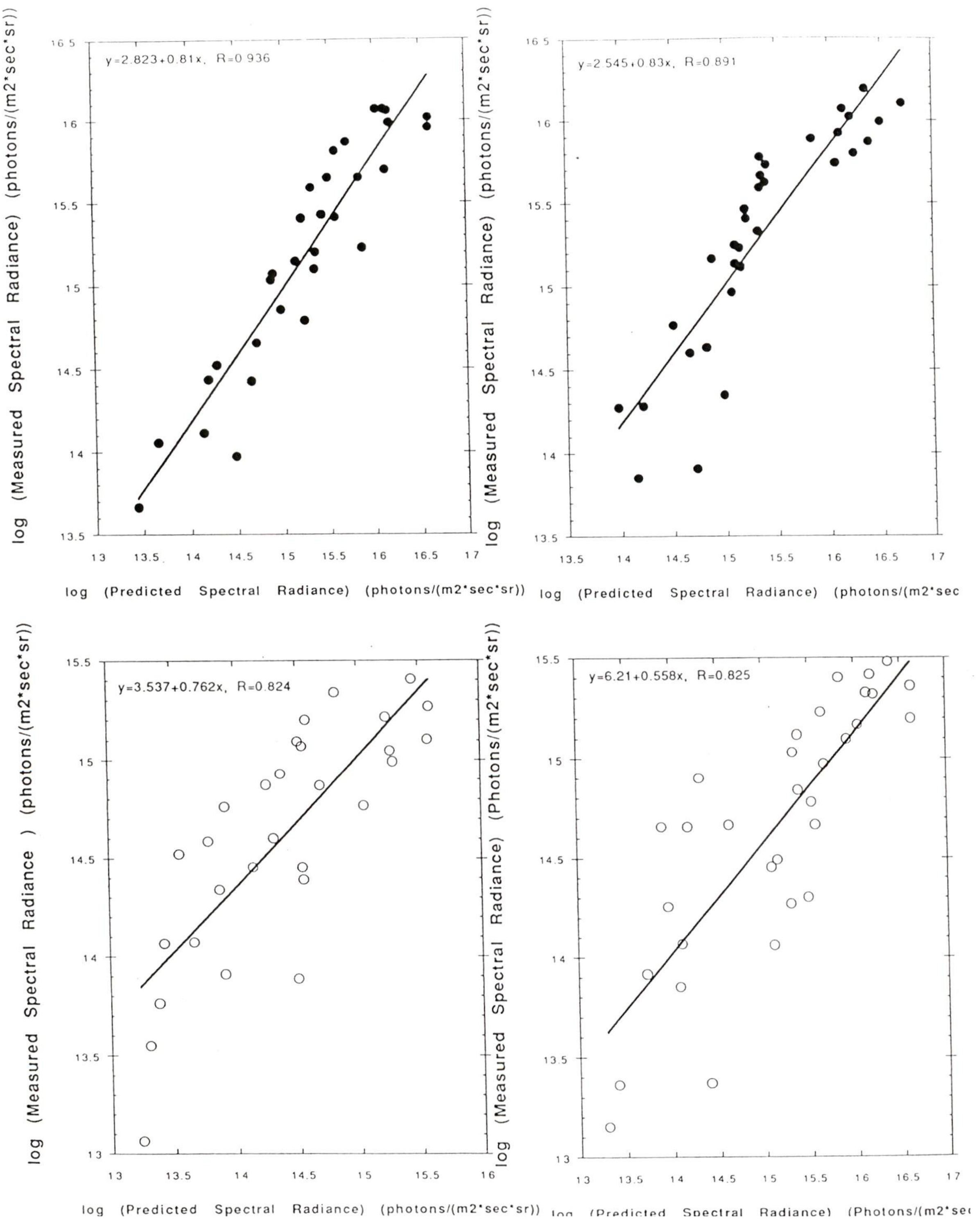


Figure 4.5. Measured vs. predicted $\log_{10}$ spectral radiances for February and March. Top two graphs are pooled data from all depths at 131° scattering angle (February at top left corner). Lower graphs show data for 49° scattering angle (bottom right graph shows data from early March). Note the close resemblance in the regression lines at each angle between months.



Discussion

Studies of light scattering under controlled experimental conditions have found results differing by a factor of at least two with predicted intensities using Mie formulae (Hepplestone and Lewis, 1968). These differences, unexplained by experimental error, have been associated with the inherent anisotropy of the particles used (Maron et al., 1963).

Predictions in natural waters are more difficult because of the additional errors brought about by the biochemical complexity of its components and by uncontrolled variables. However, improvements to the results presented in this study could be obtained by restricting the measured intensities to a finer angle (thus lowering the risk of multiple scattering) and measuring smaller size particles that may be contributing an important part of the scattered intensity through Rayleigh scattering, especially at small wavelengths. Predicted scattering intensities at angles close to 180° would fit observations better with increasing particle size.

Measurements should also be restricted to surface waters where directionality of the sun's rays is highly maintained after correction for refraction (Fig. 4.1). Wave action may induce errors in the predicted surface values due to complex patterns of refraction. This may have contributed to the discrepancies in Figure 4.4 at 0.3 m depth (see also Fig. 4.1). Measurements under calm water conditions should therefore be favoured.

A major assumption and probable cause of error in this study was the inorganic refractive index assigned to all the particles. Undetected break-down products of chlorophyll (phaeopigments) could be absorbing part of the light scattered or scattering light to a lower extent, which would invalidate this assumption. This would explain the higher values obtained for predicted UV radiances versus those measured. One way to correct for differential scattering and absorption by organic material is to assign a complex term representative of the overall absorption by cells in the water to the refractive index. The Mie calculations are far more complex in this case though, and the results may not be improved because of the independent scattering properties of inorganic vs organic particles.

The differences between observed and predicted components deviate little between the two months studied (Fig. 4.5). Because the same assumptions and conditions applied to the gathering of data from both dives, and given the similarities in phytoplankton levels between both months (Table 4.2), the results may reflect a similar ratio of inorganic to organic particulates in the lake's water. If predicted and observed results are always different by a constant factor, the ratios at different times of the year could then be correlated with biochemical measurements of organic and inorganic matter concentrations in the lake's water to act as limnological indicators of these parameters.

Although many uncontrolled variables confound results in natural waters, this study shows that the scattered light field in Lake Cowichan can be approximated, on average, within a factor of 3.76 using Mie scattering theory. The discrepancies found between predicted and measured radiances are not due to Mie theory, but rather to the simplifications made throughout the study. Clearly, scattering by water molecules and absorption by phytoplankton cells, bacteria and DOM must also be accounted for to obtain better approximations.

Chapter 5

Spectral radiance, polarization components, and salmonid vision

Introduction

Various fish species are sensitive to polarized light (halfbeak, A. Zenarchopterus, Waterman and Forward, 1972; goldfish, C. auratus, Hawryshyn and McFarland, 1987; sunfish, L. cyanellus, Cameron and Pugh, 1991; rainbow trout, O. mykiss, Hawryshyn et al., 1990). In the case of salmonids, behavioural experiments have shown that these fish are able to orient themselves to the most polarized plane of light provided UV light is part of the stimulus (Hawryshyn et al., 1990). In so doing, they require the incident light to be at least 65% polarized (Hawryshyn and Bolger, 1990)

In nature, polarized light is created by particles scattering light according to the general Mie theory (Mie, 1908), described in the previous chapter. A special case of this theory, when the scattering particles are smaller than approximately 1/10th of the incident light's wavelength, predicts 100 % polarization of the light scattered normal to the direction of the particle dipole (Van de Hulst, 1957). This ideal situation is never the case in nature; thus, animals using polarized light as a visual aid must be sensitive to lower polarization levels.

Although a series of ecological advantages have been proposed for fishes with such a sensory capability in the UV range, the UV polarized light levels in natural water bodies inhabited by these animals have never been calculated or measured. It was therefore the goal of this study to estimate such levels for Lake Cowichan. In order to do this, the radiance measurements obtained in the UV wavelengths from the previous chapter were used to calculate scattered intensity components with E-vectors perpendicular and parallel to the plane of observation. The scattered intensities and its percent polarizations were compared to similar values used in laboratory experiments eliciting fish responses (Beaudet et al., 1991; Hawryshyn et al., 1990). This enabled us to assess whether UV polarized light vision can be relevant in a mesotrophic lake.

Materials and Methods

Polarization percentage calculations

Using intensity and particle readings from the previous chapter, the polarized light components can be calculated from the following equations:

$$I_{perp.}=(I)(\partial^2 i_1/(8\pi^2))(c)\ \int \exp(-kd)dV,$$

$$I_{para.}=(I)(\partial^2 i_2/(8\pi^2))(c)\ \int \exp(-kd)dV,$$

where the variables are as described in chapter 4.

Knowing these values, percent polarizations for each wavelength were calculated in the following manner (Jerlov, 1976):

$$\%\ \text{polarization}=\ (I_{max}-I_{min})(100)/(I_{max}+I_{min}),$$

where I_{max} and I_{min} are the intensities in the maximum and minimum polarized light planes. In this study, $I_{perp.}$ and $I_{para.}$ may not represent scattered radiances in the maximum and minimum polarization planes, so the values found for this ratio may not be their maximum. In all cases, one plane was more polarized than the other, and the scattered intensity in this plane was designated I_{max}.

The previous calculations were performed for the following wavelengths: 360, 380, 400, 440 and 480 nm. The alpha band from the absorption spectrum of salmonid UV cones absorbs approximately from 300 to 450 nm; the peak absorption is centered at 380 nm as determined by Compound Action Potential recordings from the optic nerve of rainbow trout ,O. mykiss (Beaudet et al., 1991).

It is to be realized that whatever errors were made in the previous chapter in predicting radiances are also part of this chapter's calculations. However, because percent polarization is the quotient of two radiance values, the errors in previous calculations will cancel out (provided the previous correction for threshold distance as a function of wavelength is valid).

Cone aperture and retinal stimulation

The cone aperture (30°, or 1 steradian) corresponds approximately to one half of the maximum angle separating two light rays that would stimulate a specific region of the retina (Fig. 5.1). To calculate this, 10 pupil diameters from five fish averaging 18.71g in weight (s.e.= 3.61g) were averaged to give 3.84 mm (s.e.= 0.76mm). This slightly curved distance (denoted P) can be regarded as part of a circle centered at a point in the retina having radius r (Fig. 5.1, r had an average of 6.79 mm (s.e.= 0.56 mm) for the five fish). It follows that $P= r \int \sin\Omega$, where Ω is the angle between the two most distant rays that are able to reach the center of the circle, and the integration limits are from 0 to Ω. Inserting the previous averaged values into the equation and solving for Ω, this angle can be found to be equal to 63.89°. The restriction of outside light stimulation imposed by this angle may be important for the mechanics of polarization sensitivity in young salmonids (see discussion).

Results

The polarized components and calculated percent polarizations for each wavelength are shown in Table 5.1. At each depth only wavelengths with scattered radiances above 10^{-4} μmoles/(m^2*sec*sr) were used in the calculation (this value corresponds to the minimum 'available irradiance' needed for stimulation of UV cones in salmonids, Beaudet et al., 1991). The calculated values show that the light is sufficiently polarized at a scattering angle of 49°, for 440 and 480 nm, to drive salmonid visual behaviour (Hawryshyn et al.,1990). Percent polarized light values are lower in the sun direction (131° angle), results which have been shown by Timofeyeva (1969) for polarized light in the plane of the sun. In general, percent polarization decreases with depth, which may be a consequence of increased multiple scattering.

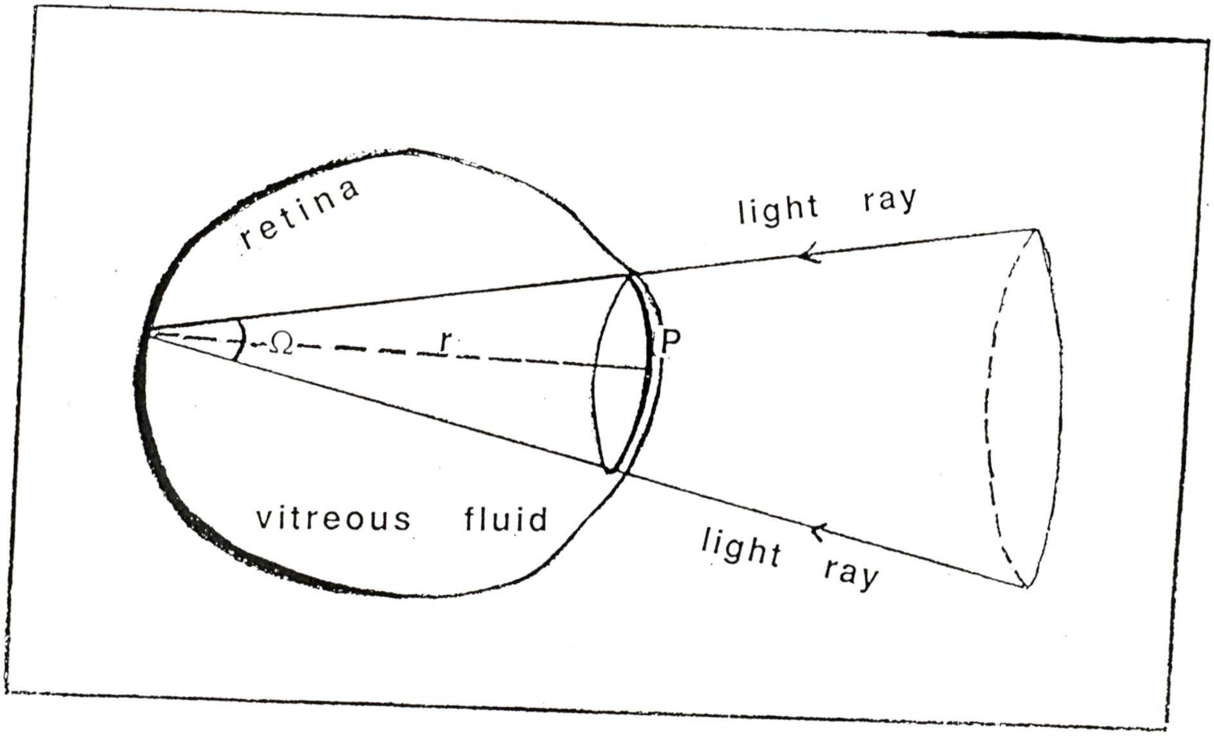
Polarization results in the plane of the sun are very different from similar results in the plane perpendicular to the sun's plane (the plane of the sun cuts the sun in two in the direction of the incident light, Timofeyeva, 1969). Percent polarization in the plane perpendicular to the sun approximates two Bell-curve distributions symmetrical with respect to the sun's plane and having maxima at 90° to it. The uniformity in horizontal polarized light visual fields with orientation along the sun's plane may be a further advantage for fish orienting in this plane, as suggested in chapter 1. A uniform background irradiance and hue distribution for both eyes may be more apt for detecting minute changes in the visual field.

Table 5.1. Predicted polarization components and polarization percentages for the various wavelengths studied as a function of time and depth. The abbreviations **s** and **as** stand for the angles 131° (horizontal sun reading) and 49° (horizontal antisun reading). All radiance values (Imax and Imin) should be mutiplied by 10^{-2} to give the predicted polarized light components in µmoles (of photons)/(m^2*sec*sr). Note that various polarization percentages at 49° are over 65%, a value which is sufficient for salmonid orientation using plane polarized light (Hawryshyn et al., 1990).

Depth (m)

February		O.3			3			6			9		
∂(nm)	°	I_{min}	I_{max}	%pol	I_{min}	I_{max}	%pol	I_{min}	I_{max}	%pol	I_{min}	I_{max}	%pol
360	s	0.009	0.017	31	-	-	-	-	-	-	-	-	-
	as	0.022	0.0302	18	-	-	-	-	-	-	-	-	-
380	s	0.111	0.177	21	-	-	-	-	-	-	-	-	-
	as	0.019	0.039	34	-	-	-	-	-	-	-	-	-
400	s	0.501	0.718	18	0.02	0.03	20	-	-	-	-	-	-
	as	0.061	0.126	35	-	-	-	-	-	-	-	-	-
440	s	0.854	1.34	22	0.163	0.196	9	0.0036	0.0039	4	-	-	-
	as	0.054	0.254	65	0.057	0.25	63	-	-	-	-	-	-
480	s	1.19	1.27	3	0.161	0.179	5	0.059	0.062	3	0.015	0.016	5
	as	0.178	0.461	44	0.0079	0.031	59	0.003	0.007	40	-	-	-
March													
360	s	0.013	0.014	1	-	-	-	-	-	-	-	-	-
	as	0.0048	0.016	54	-	-	-	-	-	-	-	-	-
380	s	0.028	0.033	7	-	-	-	-	-	-	-	-	-
	as	0.088	0.243	47	-	-	-	-	-	-	-	-	-
400	s	0.1	0.132	14	0.0054	0.0102	31	-	-	-	-	-	-
	as	0.131	0.444	54	0.0168	0.025	20	-	-	-	-	-	-
440	s	0.905	1.035	7	0.053	0.067	12	0.028	0.047	25	0.01	0.013	11
	as	0.154	1.23	78	0.034	0.171	67	0.0049	0.015	51	-	-	-
480	s	1.76	1.94	6	0.195	0.234	9	0.123	0.142	7	0.05	0.06	9
	as	0.612	2.12	55	0.103	0.31	50	0.02	0.051	43	0.004	0.01	44

Figure 5.1. Drawing of a fish's eye showing the biggest angle of incoming radiation that can stimulate a given point in the retina. Different parts of the retina will be stimulated by parts of the sky with different total % polarizations.



Discussion

The predicted percent polarizations show that fish navigation using polarized light should be possible near surface waters (Hawryshyn et al., 1990). It is important to realize that these values could be higher for the angles measured (see methods) and should be higher at an angle of 90° to the incident light (Rayleigh, 1889). Percent polarization reaches a maximum around 440 nm and seems to decrease towards longer wavelengths, as would be expected from the inverse dependence of scattered light with increasing wavelength (Rayleigh, 1889). Instead, the highly scattered short part of the UV spectrum may show percent polarizations that are close to being significant for visual processes (Table 5.1, March, 360 nm value), but they are also highly attenuated by the water. Based on these results, the hypothesis is proposed that polarized light vision in salmonids and other pelagic animals living in mesotrophic or eutrophic waters uses mainly wavelengths between 390 and 450 nm, i.e. wavelengths towards the short wavelength part of the spectrum. Interestingly, the ß absorption band of the long wavelength cone mechanism of salmonids absorbs maximally around 400 nm. It may be that this band and the long wavelength side of the UV alpha band provide the necessary information for polarized light navigation in nursery lakes and coastal areas, while the ß band of the middle wavelength mechanism (which absorbs at lower UV wavelengths) may be used with the UV alpha band for navigation in clear waters of the open ocean and for contrast enhancement in all environments. If such is the case, polarized light experiments should detect a variation in the fish's sensitivity to the plane of most polarized light when isolating the UV and long wavelength cone mechanisms and testing for wavelengths between 390 and 450 nm; shorter wavelengths are highly attenuated in the water column and therefore of limited use in meso-eutrophic waters.

The realization that each point in the retina receives input from a particular region of the outside environment at any instant in time (a cone of 63.89° approximate apex for young salmonids, Fig. 5.1) may have important implications for hypotheses regarding mechanisms behind polarization vision. Because salmonids exhibit behavioural responses only if a minimum number of photons are polarized (Hawryshyn et al., 1990), neural signals from the cones to higher nervous centers must depend on the percent polarization of the light stimulus from the portion of the outside environment illuminating them. It follows that, since each point in the retina is illuminated by regions of the environment that differ in polarization levels, comparison of neural outputs from cones in different parts of the retina would permit detection of the band of most polarized light. This, especially, if the cones are found across the retina, so that inputs from all parts of the outside environment are processed. It is worth noticing that this hypothesis for the detection of the plane of most polarized light does not require neural outputs from more than one type of cone mechanism sensitive to polarized light (Hawryshyn, 1992), nor that the cones in one section of the retina be aligned in specific directions to those in others (Cameron and Pugh, 1991). All that is required is sufficient polarized light in the environment to drive one type of cone sensitive to the polarized wavelengths and another type of cone that is insensitive to polarized light for brightness compensation (Ribi, 1980). The visual field of both types of receptors would have to be equal so that mean intensity and degree of polarization would not vary for a given region of the retina containing them (i.e. a unit in the retinal mosaic).

The above model for detection of the plane of most polarized light does not conform to Kirschfeld's (1972) model based on simultaneous inputs from three cones (of which all may be polarization sensitive or two polarization sensitive and one polarization insensitive, Wehner, 1983, 1989). Thus, an animal with only one cone type sensitive to polarized light would not be able to detect unambiguously the E-vector of the incident light, unless it sampled temporaly the outside light cue by moving its eyes (Ribi, 1980). The possibility therefore exists for such a system to be used in nature.

Behavioural and electrophysiology studies have shown that goldfish and juvenile rainbow trout can detect the E-vector of polarized light even if the animal is immobile (Hawryshyn and McFarland, 1987; Parkyn and Hawryshyn, In Press). It would therefore seem that for these fish species, polarization detection based on temporal displacement of one cone mechanism would be redundant. However, the observed loss of cones during development of rainbow trout may suggest that a one cone mechanism could be active.

Large rainbow trout (>30g) lose their accessory corner cones (believed to be the carriers of UV opsin) at all sites except along the Embryonic Fissure and periphery of the retina (Beaudet et al., In

Press). It is interesting to note at this point that the Embryonic Fissure runs from the optic nerve head anteriorly, to the periphery of the retina, in a curvilinear fashion. It is also interesting to note that it is during this time in the fish's life history (open ocean dwelling for various Pacific salmonids) that navigation by polarized light would be most beneficial as there are few other cues in the environment to be guided by. A line of UV receptors (such as that found along the Embryonic Fissure) is all that is needed geometrically to detect a portion of the band of most polarized light using only the UV receptor. Electrophysiology studies on the other hand have shown that large rainbow trout (<30g) are insensitive to the vertical E-vector of polarized UV light, and this has been linked to the loss of UV cones from the retina (Parkyn and Hawryshyn, In Press). Could it be possible that adult rainbow trout are merely detecting differences in percent polarization using one type of polarization sensitive photoreceptor (either the UV cones along the Embryonic Fissure or the double cones in the rest of the retina, along with short wavelength sensitive cones) to guide their movements? This would also explain the inability of large rainbow trout to orient during behavioural experiments (Hawryshyn et al., 1990). Alternatively, the Compound Action Potential technique used to record from the ganglion cells used by Parkyn and Hawryshyn (In Press) may not be sufficiently sensitive to detect polarization responses in large fish (Coughlin and Hawryshyn, In Press).

Hawryshyn et al. (1990) showed that when illuminated by polarized light of wavelengths longer than 450 nm, young salmonids were less able to orient to the E-vector of polarized light, than when illuminated by wavelengths smaller than 450 nm. However, in these experiments, none of the cone mechanisms were adapted prior to stimulation with UV polarized light. The results therefore do not show that the observed fish behaviour could not be achieved with only one cone mechanism sensitive to the UV polarized wavelengths. Furthermore, because of the different scattering patterns, produced by water molecules and dissolved "Rayleigh-type" particles, with E-vector direction, it is conceivable that the fish were orienting to differences in intensity distributions impinging on non-ventral parts of the retina (i.e. receiving scattered light). This possibility should also be evaluated in future research. Finally, the fact that 3 out of 11 fish "statistically" oriented to the E-vector under non UV conditions leaves unanswered questions to the use of other wavelengths for orientation (it is to be noted that this ratio represents more than 25% of the sample size tested). Behaviour orientation experiments of the type performed by Hawryshyn et al. (1990) under a middle and long wavelength adapting background should determine whether two cone outputs are needed for salmonid orientation to plane polarized light.

Although there is evidence to support a polarized light mechanism based on two inputs (a member of the double cone and the UV cone) for rainbow trout (Hawryshyn, 1992), further research needs to erase other possibilities while continuing to assert this model. One may also ask from an evolutionary perspective what are the advantages of investing in a two polarized light sensitive input system when a one input system (and a brightness detector) may "theoretically" do the job for orientation and contrast enhancement.

From a biophysical perspective, a structural mechanism must also be found that would explain laboratory observations. With respect to this issue, structures such as the cristae of mitochondria occurring in the ellipsoids of cones may be aligning in a particular direction and acting as polarizers. Other structural elements present in great quantities in retinal tissue such as microtubules (Matesic et al., 1992) could also form polarizing filters. Yet other mechanisms for creating polarized light like interference reflectors and wave-guides should also be considered.

Chapter 6

Photopigment analysis of spectral sensitivity curves from light-adapted rainbow trout and goldfish

Introduction

Colour vision in vertebrates is a result of differential excitation of retinal photoreceptors absorbing at common wavelengths. Under a white light background of moderate intensity (i.e. a background within the active range of intensity for operation of all pigments), the resulting spectral sensitivity of the animal will be a function of the absorbance of all pigments and neural interactions taking place at the photoreceptor level (Scholes, 1975; Normann et al., 1984) and at higher processing layers in the retina and the brain (Neumeyer, 1984). Pigment absorption spectra (obtained from microspectrophotometric measurements, Liebman and Entine, 1964, Harosi, 1976) have been used to model spectral sensitivity curves of isolated cone mechanisms (DeMarco and Powers, 1991), zones of interaction between mechanisms (Yager, 1969, Normann et al., 1984; Neumeyer, 1984, De Marco and Powers, 1991) and "full-spectrum" (400-750 nm) spectral sensitivity curves from light-adapted animals (Yager, 1967, Powers, 1978, DeMarco and Powers, 1991). The main results from these studies show that the spectral sensitivity curves obtained under white adaptation conditions can be approximated by linear models of the form:

$R= K_1\ (A_S) + K_2\ (A_M) + K_3\ (A_L)$ (Yager, 1967, 1969)

$R= K_1\ (A_S) + K_2\ (A_M) - K_3\ (A_L)$ (Powers, 1978)

where R= spectral sensitivity response,

$K_{1,2,3}$= coupling coefficients, and

$A_{S,M,L}$= absorbance of the short, middle and long wavelength cones.

Although these models may conform to the general shape of spectral sensitivity curves in general, they are not very accurate in regions of intensive cone interaction. In such regions, substractive models have been used to model the interaction between the two highest absorbing pigments in the region of the spectrum examined (Naka and Rushton, 1966, Sperling and Harwerth, 1971, Neumeyer, 1984). However, in such studies, the contribution of a photopigment to the spectral sensitivity curve in different regions of the spectrum is often assigned opposite signs to match the data, which is counter-intuitive to a single and consistent function of each type of photoreceptor accross the spectrum (Sperling and Harwerth, 1971, Hawryshyn and Harosi, 1991). Furthermore, such models, although accurately fitting the response in the cone interaction region, may not be accurate in the remaining region of the action spectrum studied (see Hawryshyn and Harosi, 1991).

In a theoretical treatment of photopigments, Sirovich and Abramov (1977) showed that any "pseudopigment" curve had to conform to the equation:

$$R=(\Sigma\ \beta_i\ A^p{}_i(f))^{1/p} \qquad (1)$$

where R is the response curve, $A_i(f)$ is the absorbance of pigment i at light of frequency f, and p is an exponent resulting from the mathematical requirement that the function describing the spectral sensitivity curve is differentiable at the origin (see Sirovich and Abramov, 1977). This last requirement would not be met if the curve were a "spike" (i.e. the response was independent of wavelength); given the shape of spectral sensitivity curves, this is not a rigorous restriction. The β_i coefficients are the differential coefficients resulting from the previous requirement, they are the coupling coefficients derived from the best fit of the model to the data. Equation 1 is non linear. The coupling coefficients and the exponent p describe the contribution of each photopigment to the spectral sensitivity curve (which can be regarded as a combination of "pseudopigments") and the complex interactions occurring between cones and at higher processing levels.

In order to model the spectral sensitivity curves of light adapted goldfish and rainbow trout, equation 1 was inserted into a computer program that used the Simplex algorithm (Caceci and Cacheris, 1984). The coupling coefficients generated by the model were analyzed in terms of the intensity of the background adaptation used. The results showed the contribution of each cone type to the visual response with varying radiance.

Materials and methods

Spectral sensitivity curves

The data for the goldfish curves were obtained from Hawryshyn (1991). The spectral sensitivity values were normalized to expand the same range as the absorption curves for the pigments. These were generated from Bernard's (1987, pers. comm.) eighth order polynomial template for vertebrate cone absorption.

The measurements of spectral sensitivity for light-adapted rainbow trout were obtained using compound action potential recordings from the optic nerve of young rainbow trout (<15 g in weight). The surgical and recording procedure used was similar to the one appearing in DeMarco and Powers (1991); however, platinum electrodes were inserted dorso-laterally forward through the optic tectum into the optic nerve, instead of using a suction electrode in which to draw the optic nerve. Also a third order polynomial was fitted to the plot of log response amplitude vs. wavelength (Beaudet et al., 1991). A third order fit is more accurate than the first degree fit used by DeMarco and Powers (1991).

Curve-fitting

The Simplex algorithm optimizes the model fit to the data by minimizing the sum of the squared residuals: $\Sigma\ (r_i-r_i')^2$, where the r_i are the spectral sensitivity response values observed, and the r_i' refer to the predicted response values according to equation 1. This optimization is achieved by assigning weights to each residual square $(r_i-r_i')^2$ so that the sum of square residuals (depicting the entire spectral sensitivity curve fit) is a minimum. To do this, the algorithm creates a 6th dimensional simplex (a simplex is a geometric figure with one more vertex than the space in which it is defined has dimensions) and "moves" it through the data points according to four mathematical mechanisms (reflection, expansion, contraction and shrinkage) (Nedler and Mead, 1965, Caceci and Cacheris, 1984). The coupling coefficients and exponent (p) were obtained using this technique.

Results

The fits of equation 1 to the rainbow trout and goldfish data are very good (Figs. 6.1 and 6.2); in every case the sums of squares (SS) is less than 1.04. Because the goldfish data was obtained using a behavioural technique, as opposed to the electrophysiology procedure used for obtaining the rainbow trout data, the scatter is somewhat higher for goldfish data.

The coefficients in Figure 6.1 are highest for the UV and short wavelength cones, which is consistent with the two peaks in sensitivity shown on the curve. Similarly, the decrease in sensitivity in the middle and long wavelength parts of the spectrum is depicted by the smaller coefficients found for the highest absorbing cones in these parts of the "visual" spectrum. The model curve is also seen to accurately follow depressions in the spectral sensitivity curve where antagonistic interactions between cones have been postulated by various authors (Powers and Easter, 1978, Hope and Dawson, 1979, Neumeyer, 1984).

Analysis of the goldfish data using equation 1 shows that the UV cones are most sensitive at intermediate intensities (UV=0.92, curve 3, Fig. 6.2). The same conclusion had been reached by Hawryshyn (1991) matching nomograms to the data. However, the contribution of each photopigment was unknown. In these series of curves, the long wavelength cones appear to dominate the spectral sensitivity curves at all times. This may result from the fact that the long wavelength cone expands the broadest action spectrum and can therefore dominate in photon catch.

It is interesting to note that intermediate intensities correspond to light conditions during crepuscular periods (Hawryshyn, 1991), a time when polarization of UV wavelengths in the sky may be a navigational tool used by salmonids (Hawryshyn, 1992).

Figure 6.1. Characteristic spectral sensitivity curve from light-adapted rainbow trout (SS=0.011). The coupling coefficients are indicated on top of the figure.

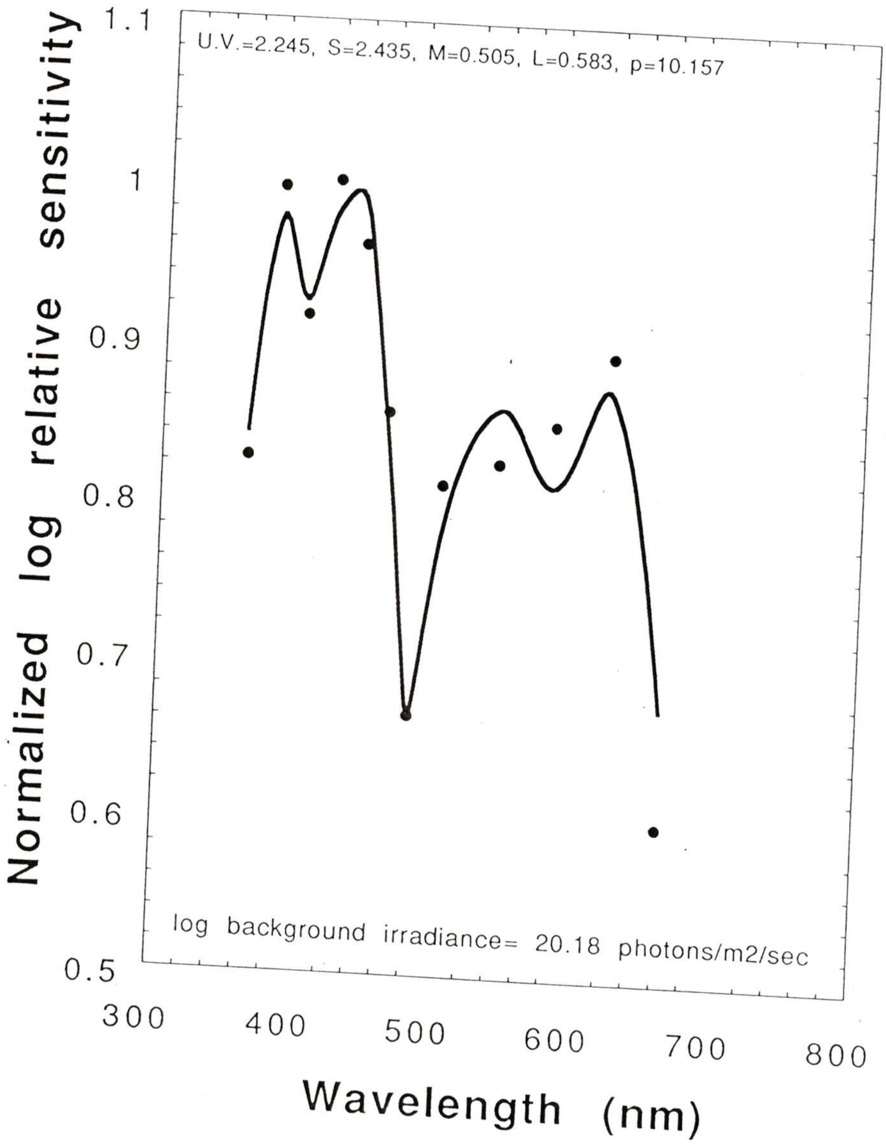
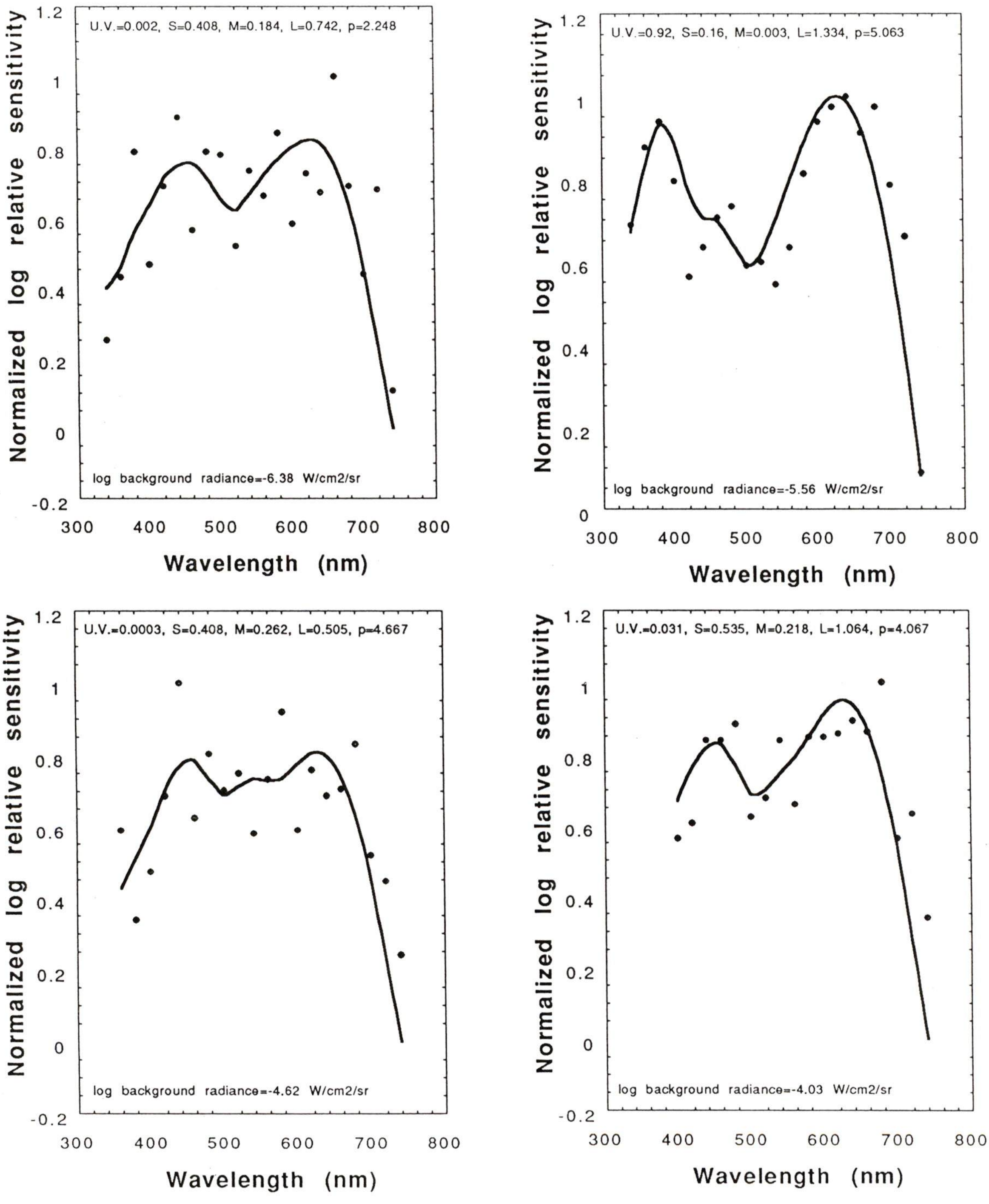


Figure 6.2. Spectral sensitivities of goldfish under backgrounds of varying light intensity. Data points from Hawryshyn (1991).



Discussion

The accuracy of the fits obtained using equation 1 shows that this non linear summation model describes best the cone mechanism interactions making up the spectral sensitivity curves. These are observed to be antagonistic as described by various authors (Newmeyer, 1984). Linear models were also attempted, but the fits using these models were less satisfactory. The non linearity of equation 1, and the fact that the p parameter does not appear to follow a smooth trend with increasing wavelength (Fig. 6.2), suggests that complex signal interactions are taking place to make up the signal recorded from the optic nerve. Further research using more accurate electrophysiological recordings should determine if this is an artifact of the scatter in the data of Figure 6.2, or whether the parameters in equation 1 are indeed unpredictable. Whatever the case, Equation 1, or any other curve-fitting model, cannot reveal the visual processing taking place. Only a thorough understanding of the signal processing at every level from the photoreceptor cell layer to the optic tectum, would permit an accurate prediction of the spectral sensitivity given an incident light stimulus.

Chapter 7

General Discussion

The light measurements presented in this work clearly show that in meso-eutrophic waters, all the photoreceptor cones can be stimulated to significant depths (15 meters or more). Thus, a wide arrangement of visually-mediated behaviours are possible provided the animal has the retinal capabilities of processing the light cues.

The presence of UV light in surface waters indicates that UV sensitivity observed in laboratory conditions (Browman et al., 1992, Hawryshyn et al., 1990) may be relevant in nature. Whether this is the case will be extremely difficult to show, the reason being the impossibility of dissociating visual light cues from all other cues (magnetic, chemical, thermal and salinity dependent, Quinn, 1980, Westerberg, 1982, Quinn and Dittman, 1991) present in nature. Perhaps statistical treatment of data from tagged salmon wearing non-UV-transmissive lenses (or some other mechanism to prevent the use of UV light) would show that UV visual cues play a significant role in salmon feeding performance and/or orientation during migration. In any case, large scale experiments in natural conditions are required to test whether the postulated behaviours are practiced by wild salmon.

One of the subjects presented in this work that needs field measurements is polarized light. To date, there are no measurements of UV polarized light, yet the entire orientation hypothesis relies on such visual cues being present in nature. I suspect, from the theoretical results presented in chapter 5 that sufficient polarized UV light for visual cues will be restricted to near surface depths (less than 9 m) for meso-eutrophic waters. However, because of the many assumptions made in these calculations, only field measurements will show exactly what is available for visual processing.

Perhaps one of the interesting avenues for further research is work in the open ocean, because transmission of UV wavelengths would be more favourable in this environment (Smith and Baker, 1979). The overwhelming presence of small ("Rayleigh-type") particles in this environment would further increase the polarization of UV light and, as a consequence, the chances of it being used by salmon. Topographically, the absence of land marks and derived cues from river outflows (such as salinity gradients and chemical odours), would favour the use of other senses (such as polarized light navigation) by adult salmonids returning from the ocean to their home streams. Interestingly, as outlined in chapter 5, adult salmonids retain corner cones along the Embryonic Fissure (Kunz, 1987, Beaudet et al., In Press), which may be functional during home migration.

The detection of UV light by animals living in coastal waters of Vancouver Island, where penetration of this light is considered low, suggests that this sensory capability may be wide spread in tropical oceans where waters are oligotrophic (Jerlov, 1976). The great diversity in colours of corals and reefs in these environments may be appropriate for extensive use of UV light (reflection off sands and corals, contrast enhancement against bright colours..etc). Temperatures and sunlight conditions are also favourable to researchers studying UV light!

The data collected in this project will hopefully serve not only vision researchers but also the entire community of marine biologists, oceanographers and meteorologists working with some aspect of light. For instance, Photosynthetic Available Radiation (PAR) can be calculated using the values presented. The light values can also be used as accurate indicators of the turbidity of meso-eutrophic waters. Finally, in an era when ozone depletion may have a significant impact on biological life, these values may be used along with others to detect possible impacts on aquatic ecosystems.

Literature cited

AVERY, J.A., BOWMAKER, J.K., DJAMGOZ, M.B.A., and DOWNING, J.E.G. (1983). Ultraviolet sensitive receptors in freshwater fish. Journal of Physiology (London) 334: p. 23.

BAKER, K.S. and SMITH, R.C. (1982). Spectral irradiance penetration in natural waters. In The role of solar ultraviolet radiation in marine ecosystems. (Edited by J. Calkins), Plenum Publishing Corporation pp. 233-245.

BEATTY, D.D. (1966). A study of the succession of visual pigments in Pacific Salmon (Oncorhyncus). Canadian Journal of Zoology 44: 429-455.

BEAUCHAMP, R.D. and LOVASIK, J.V. (1973). Blue mechanism response of single goldfish optic fibers. Journal of Neurophysiology 36: 925-939.

BEAUDET, L., BROWMAN, H.I. and HAWRYSHYN, C.W. (1991). Ontogenetic loss of U.V. photosensitivity in rainbow trout, determined using optic nerve compound action potential recording. Society of Neuroscience Abstract 17(1): 299.

BEAUDET, L., BROWMAN, H.I, and HAWRYSHYN, C.W. (1992). Developmental changes in the spectral sensitivity of rainbow trout optic nerve fibers. Vision Research (In Press).

BERNARD, G.D. (1987). Spectral characteristics of butterfly L-receptors using extended Dartnall/McNichol template functions. Journal of the Optical Society of America 2 (13): 123.

BIGGS, W.W. (1984). LI-1800UW underwater spectroradiometer instruction manual. Publication No. 8405-0037. Li-Cor Inc. pp. 126.

BOWLING and TYLER, (1986). The underwater light-field of lakes with marked physico-chemical and biotic diversity in the water column.

Journal of Plankton Research. 8: 69-77.

BOWMAKER, J.K. and KUNZ, Y.L. (1987). Ultraviolet receptors, tetrachromatic colour vision and retinal mosaics in the brown trout (Salmo trutta): Age dependent changes. Vision Research 27: 2101-2108.

BRIDGES, C.D.B. (1972). The rhodopsin-porphyropsin visual system. p. 417-480. In H.J.A. Dartnall (ed.) Photochemistry of vision. Springer-Verlag Berlin-Heidelberg-New York.

BROWMAN H.I. and HAWRYSHYN, C.W. (1992). Thyroxine induces a precocial loss of ultraviolet photosensitivity in rainbow trout (Oncorhyncus mykiss, Teleostei). Vision Research 32: 2303-2312.

BROWMAN, H.I., NOVALES-FLAMARIQUE, I. and HAWRYSHYN, C.W. (1992). Ultraviolet photoreception, prey-search behaviour, and visual acuity in juvenile zooplanktivorous fish. American Zoologist abstract 45 (3): 2.

BROWN, R.M. and BORSTAD, G.A. (1981). Canada-France ocean optics experiment: sea-truth data for stations in British Columbia waters. DSS file No. 6-1850-000-000-130. Department of Fisheries and Oceans, Sidney, British Columbia.

CACECI, M.S. and CACHERIS, W.P. (1984). Fitting curves to data. Byte 5: 340-360.

CAMERON, D.A. and PUGH, E.N. (1991). Double cones as a basis for a new type of polarization vision in vertebrates. Nature 353: 161-164.

CHEN, H. and NAGARAJA RAO, C.R. (1968). Polarization of light on reflection by some natural surfaces. Brittish Journal of Applied Physics 2 (1): 1191-1200.

COUGHLIN, D.J. and HAWRYSHYN, C.W. (1993). Ultraviolet sensitivity in the torus semicircularis of rainbow trout (Oncorhyncus mykiss). Vision Research (In Press).

DAVIES-COLLEY, R.J., VANT, W.N. and WILCOCK, R.J. (1988). Lake water color: Comparison of direct observations with underwater spectral irradiance. Water Research Bulletin American Water Resources Association. 24 (1): 11-18.

DeMARCO, P. J. Jr. and POWERS, M.K. (1991). Spectral sensitivity of ON and OFF responses from the optic nerve of goldfish. Visual Neuroscience 6: 207-217.

DENTON, E.J. and NICOL, J.A.C. (1965). Polarization of light reflected from the silvery exterior of the bleak, Alburnus alburnus. Journal of marine biology Association of the U.K. 45: 705-709.

DILL, P.A. (1971). Perception of polarized light by yearling Sockeye salmon (Oncorhyncus nerka). Journal of the Fisheries Research Board of Canada 28: 1319-1332.

DUNTLEY, S.Q. (1963). Light in the sea. Journal of the Optical Society of America 53: 214-233.

EINSTEIN, A. (1910). Theorie der Opaleszenz von homogenen Flussigkeiten und Flussigkeitsgemischen in der Nahe des kritischen Zustandes. Annual der Physik 33: 1275-1298.

FRENCH, R., BILTON, H., OSAKO, M., and HARTT, A. (1976). Distribution and origin of sockeye salmon (Oncorhyncus nerka) in offshore waters of the north Pacific ocean. International North Pacific Fisheries Communication Bulletin 34: 113 pp.

GOWER, J.F.R. and BORSTAD, G.A. 1990. Mapping of phytoplankton by solar-stimulated fluorescence using an imaging spectrometer. International Journal of Remote Sensing 11: 313-320.

GROOT, C. (1965). On the orientation of young Sockeye salmon (Oncorhyncus nerka) during seaward migration out of lakes. Behaviour (Supplement). 14, 198 p.

GROOT, C. (1972). Migration of yearling Sockeye salmon (Oncorhyncus nerka) as determined by time-lapse photography of sonar observation. Journal of the Fisheries Research Board of Canada 29: 1431-1444.

HAROSI, F.I. (1975). Absorption spectra and linear dichroism of some amphibean photoreceptors. Journal of General Physiology 66: 357-382.

HAROSI, F.I. (1976). Spectral relations of cone pigments in goldfish. Journal of General Physiology 68: 65-80.

HAWRYSHYN, C.W. (1992). Polarization vision in fish. American Scientist 80: 164-175.

HAWRYSHYN, C.W. (1991). Light-adaptation properties of the ultraviolet-sensitive cone mechanism in comparison to the other receptor mechanisms of goldfish. Visual Neuroscience 6: 293-301.

HAWRYSHYN, C.W. and McFARLAND, W.N. (1987). Cone photoreceptor mechanisms and the detection of polarized light in fish. Journal of Comparative Physiology A. 160: 459-465.

HAWRYSHYN, C.W. and HAROSI, F.I. (1991). Ultraviolet photoreception in carp: microspectrophotometry and behaviorally determined action spectra. Vision Research 31: 567-576.

HAWRYSHYN, C.W., ARNOLD, M.G., CHIASSON, D. and MARTIN, P.C. (1989). The ontogeny of ultraviolet photosensitivity in rainbow trout (Salmo gairdneri). Visual Neuroscience 2: 247-254.

HAWRYSHYN, C.W. and BOLGER, A.E. (1990). Spatial orientation of trout to partially polarized light. Journal of Comparative Physiology. 167: 691-697.

HAWRYSHYN, C.W., ARNOLD, M.G., BOWERING, E. and COLE, R.L. (1990). Spatial orientation of rainbow trout to plane polarized light: The ontogeny of E-vector discrimination and spectral sensitivity characteristics. Journal of Comparative Physiology A 166: 565-574.

HAWRYSHYN, C.W. and BEAUCHAMP, R. (1985). Ultraviolet photosensitivity in goldfish: an independent U.V. retinal mechanism. Vision Research 25: 11-20.

HEALEY, M.C. (1980). The ecology of juvenile salmon in Georgia Strait, British Columbia, p. 203-229. In: W. J. McNeil and D.C. Himsworth (eds.). Salmonid ecosystems of the North Pacific. Oregon State University Press, Corvallis, OR.

HEINERMANN, P.H. and ALI, M.A. (1985). Correlation between the photic environment and porphyropsin in the Cutlips Minnow, *Exoglossum maxillingua.* Naturwissenschaften 72: 488-489.

HEINERMANN, P.H. and ALI, M.A. (1989). The photic environment and scotopic visual pigments of the creek chub, *Semotilus atromaculatus* and white sucker, *Catostomus commersoni*. Journal of Comparative Physiology A 164: 707-716.

HEINERMAN, P.H. and ALI, M.A. (1988). Seasonal changes in the underwater light climate of two Canadian shield lakes. Hydrobiologia 169: 107-121.

HECHT, E. and ZAJAC, A. (1974). Optics. Addison-Wesley Pub. Co. Reading, Mass.

HELLER, W., and TABIBIAN, R. (1962). Experimental investigations on the light scattering of colloidal spheres.IV. Scattering ratio. Jounal of Physical Chemistry 66: 2059-2066.

HEPPLESTONE, G.W. and LEWIS, P.C. (1968). Experimental observations on the angular distribution of scattered radiation from suspensions containing particles of a size comparable with the wavelength. Brittish Journal of Applied Physics 2(1): 199-206.

HOBSON, E.S. (1972). Activity of Hawaiian reef fishes during the evening and morning transitions between daylight and darkness. Fish. Bull. Nat. Mar. Fish. 70: 715-740.

HOPE, G.M. and DAWSON, W.W. (1979). Goldfish retina: electroretinographic evidence for receptor interaction. Brain Research 175: 135-139.

JEFFREY, S.W. and HUMPHREY, G.F. (1975). New spectrophotometric equations for determining chlorophylls a, b, c^1 and c^2 in higher plants, algae and natural phytoplankton. Biochemistry Physiology Pflanz. 167: 191-194.

JERLOV, N.G. (1976). Marine Optics. In Elsevier Oceanography Series, vol.14. Elsevier Science Publishing Company.

JOHNSON, W.E. and GROOT, C. (1963). Observations on the migration of young sockeye salmon (Oncorhyncus nerka) through a large, complex lake system. Journal of the Fisheries Research Board of Canada 20 (4): 919-934.

KIRK, J.T.O. (1983). Light and photosynthesis in aquatic ecosystems. Cambridge University Press, Cambridge. pp. 401.

KULLENBERG, G. (1968). Scattering of light by Sargasso sea water. Deep-Sea Research 15: 423-432.

KULLENBERG, G. (1974). Observed and computed scattering functions. In: N. Jerlov and E. Steemann Nielsen (Editors), Optical Aspects of Oceanography. Academic Press, New York, N.Y., pp. 25-49.

KUNZ, Y.W. (1987). Tracts of putative UV receptors in the retina of the two-year-old brown trout (Salmo trutta) and the Atlantic salmon (Salmo salar). Experientia 43: 1202-1204.

LE GRAND, Y. (1939). La penetration de la lumiere dans la mer. Annuels de l'Institut Oceanographique. 19: 393-436.

LENOBLE, J. (1956). Etude de la penetration de l'ultraviolet dans la mer. Nouvelles mesures. Annuels de Geophysique 12: 16-31.

LIEBMAN, P. and ENTINE, G. (1964). Sensitive low-light level microspectrophotometer: Detection of photosensitive pigments of retinal cones. Journal of the Optical Society of America 54: 1451-1459.

LOEW, E.R. and McFARLAND, W.N. (1990). The underwater visual environment. In Vision in Fishes (ed. R. Douglas and M.B.A. Djamgoz), pp. 1-43. Chapman and Hall, London.

LOEW, E.R. and WAHL, C.W. (1991). A short wavelength sensitive cone in juvenile yellow perch, *Perca flavescens.* Vision Research 31: 353-360.

LYTHGOE, J.N. (1968). Visual pigments and visual range underwater. Vision Research 8: 997-1012.

LYTHGOE, J.N. (1979). The ecology of vision. Oxford Press. New York. pp. 244.

MACKAS, D.L. (1992). Seasonal cycle of zooplankton off Southwestern British Columbia: 1979-89. Canadian Journal of Fisheries and Aquatic Sciences 49: 903-921.

MARON, S.M., ELDER, M.E. and PIERCE, P.E. (1963). Determination of latex particle size by light scattering. Journal of Colloidal Sciences18: 733-743.

MATESIC, D.F., PHILP, N.J., MURRAY, J.M. and LIEBMAN, P. (1992). Tubulin in bovine retinal rod outer segments. Journal of Cell Science 103: 157-166.

MIE, G. (1908). Beitrage zur Optik truber Medien, speziell kolloidalen Metal-losungen. Annual der Physik 25: 377.

MOREL, A. (1973). Diffusion de la lumiere par les eaux de mer. Resultats experimentaux et approche theorique. In: Optics of the Sea.

McFARLAND, W. N. (1991). Light in the Sea: The Optical World of Elasmobranchs. Journal of Experimental Zoology (Supplement) 5: 3-12.

McFARLAND, W.N. and MUNZ, F.W. (1975a). The evolution of photopic visual pigments in fishes. Vis. Res. 15: 1071-1080.

McFARLAND, W.N. and MUNZ, F.W. (1975b). The visible spectrum during twilight and its implications to vision. In Light as an Ecological Factor, vol. 2 (ed. G.C. Evans, Rockham, O. and R. Bainbridge), pp. 240-270. Blackwell, Oxford.

McFARLAND, W.N. and MUNZ, F.W. (1975c). Part II: The photic environment of clear tropical seas during the day. Vision Research 15: 1063-1070.

MOREL, A. (1965). Resultats experimentaux concernant la penetration de la lumiere du jour dans les eaux Mediterraneennes. Cahier Oceanographique17:177-184.

MUNZ, F.W. and McFARLAND, W.N. (1973). The significance of spectral position in the rhodopsins of tropical marine fishes. Vision Research 13:1829-1874.

MUNTZ, W.R.A. and A.W. WAINWRIGHT. 1978. Annual cycles in the light environments and visual mechanisms of fishes. p. 105-129. In J.E. Thorpe (ed.) Rythmic activity of fishes. Academic Press, New York.

NAKA, K.I. and RUSHTON, W.A.H. (1966). S-potentials from colour units in the retina of fish (Cyprinidae). Journal of Physiology 185: 536-555.

NATIONAL BUREAU OF STANDARDS (1949). Tables of Scattering

Functions for Spherical Particles. U.S. Dept. of Commerce, U.S. Government Printing Office, Washington, D.C.

NEDLER, J.A. and MEAD, R. (1965). A Simplex method for function minimization. Computer Journal 7: 308-311.

NEUMEYER, C. (1984). On spectral sensitivity of the goldfish. Evidence for neural interactions between different "cone mechanisms". Vision Research10: 1223-1231.

NORMANN, R.A., PERLMAN, I., KOLB, H., JONES, J. and DALY, S.J. (1984). Direct excitatory interactions between cones of different spectral types in the turtle retina. Science 224: 625-627.

OCHAKOVSKY, Y. E. (1966). On the comparison of measured and calculated scattering indicatrices of sea water. U.S. Deptartment of Commerce, Joint Publication Research Series, Report 38(816):98-105.

PAN, D., GOWER, J.F.R. and BORSTAD, G.A. (1988). Seasonal variation of the surface chlorophyll distribution along the British Columbia coast as shown by CZCS satellite imagery. Limnology and Oceanography 33: 227-244.

PARKYN, D.C. and HAWRYSHYN, C.W. (1993). Polarized light sensitivity in rainbow trout (Oncorhyncus mykiss): characterization from multiunit ganglion cell responses in the optic nerve fibres. Journal of Comparative Physiology A. (In Press).

PELLA, J.J. (1968). Distribution and growth of sockeye salmon fry in Lake Aleknagik, Alaska, during the summer of 1962, p. 45-111. In: R.L. Burgner (ed.). Further studies of Alaska sockeye salmon. University of Washington Publications Fisheries New Series 3.

PAVLOV, V.M. and GRECHUSHNIKOV, B.N. (1966). Some aspects of the theory of daylight polarization in the sea. U.S. Deptartmet of

Commerce Joint Publication Research Series, Report, 36(816): 25-44.

POWERS, M.K. (1978). Light-adapted spectral sensitivity of the goldfish: a reflex measure. Vision Research 18: 1131-1136.

POWERS, M.K. and EASTER, S.S Jr.(1978). Absolute visual sensitivity of the goldfish. Vision Research 18:1137-1147

PREZELIN, B.B., TILZER, M.M., SCHOFIELD, O. and HAESE, C. (1991). The control of the production process of phytoplankton by the physical structure of the aquatic environment with special reference to its optical properties. Aquatic Science 53 (2/3):136-186.

QUINN, T.P. (1980). Evidence for celestial and magnetic compass orientation in lake migrating Sockeye salmon fry. Journal of Comparative Physiology137: 243-248.

QUINN, T.P. (1990). Current controversies in the study of salmon homing. Ethology, Ecology and Evolution 2: 49-63.

QUINN, T.P. and DITTMAN, A.H. (1990). Pacific salmon migrations and homing: mechanisms and adaptive significance. TREE 5: 174-177.

RAYLEIGH, L. (1889). On the transmission of light through an atmosphere containing many small particles in suspension, and on the origin of the blue of the sky. Phil. Mag. 47: 375-384.

RIBI, W.A. (1980). New aspects of polarized light detection in the bee in view of non-twisting rhabdomeric structures. Journal of Comparative Physiology 137: 281-285.

RUGGERONE, G.T, QUINN, T.P., McGREGOR, I.A. and WILKINSON, T.D. (1990). Horizontal and vertical movements of adult steelhead trout, _Oncorhyncus mykiss_, in the Dean and Fisher channels, British Columbia. Canadian Journal of Fisheries and Aquatic Sciences 47:

1963-1969.

SAS Statistics Package (version 5). SAS Institute Inc. Box 8000, Cary, N.C. 27511-8000.

SASAKI, T., WATANABE, S., OSHIBA, G., OKAMI, N. and KAJIHARA, M. (1962). On the instrument for measuring angular distribution of underwater radiance. Bulletin of the Japanese Society of Scientific Fisheries 28: 489-496.

SCHOLES, J.H. (1975). Colour receptors, and their synaptic connections, in the retina of a cyprinid fish. Phil. Trans. R. Soc. Lond. B270: 61-118

SINGARAJAH, K.V. and HAROSI, F.I. (1992). Visual cells and pigments in a demersal fish, the black sea bass (*Centropristis striata*). Biological Bulletin 182: 135-144.

SIROVICH, L. and ABRAMOV, I. (1977). Photopigments and pseudopigments. Vision Research 17: 5-16.

SMITH, R.C. and BAKER, K.S. (1979). Penetration of U.V.-B and biologically effective dose-rates in natural waters. Photochemistry Photobiology 29: 311- 323.

SMITH, R.C, TYLER, J.E. and GOLDMAN, C.R. (1973). Optical properties and color of Lake Tahoe and Crater Lake. Limnology and Oceanography 18(2): 189-199.

SMOLUCHOWSKI, M.V. (1908). Molekular-kinetische Theorie der Opaleszenz von Gasen in kritischen Zustande, sowie einiger verwandter Erscheinungen. Annual der Physik 25: 205-221.

SPENCE, D.H.N., CAMPBELL, R.M. and CHYSTAL, J. (1991). Spectral intensity in some Scottish freshwater lochs. Freshwater Biology 1: 321-337.

SPERLING, H.G. and HARWERTH, R.S. (1971). Red-green cone interactions in the increment-threshold spectral sensitivity of primates. Science

172: 180-184.

SPIELHAUS, A.F. (1965). Observations of light in sea water. Office Naval Re., Washington, D.C., Dept. Geol. Geophys., Mass. Inst. Technol., Cambridge, Mass., 242 pp.

TIMOFEYEVA, V.A. (1969). Plane of vibrations of polarized light in turbid media. Izv., Atmospheric and Oceanic Physics 5 (10): 1049-1057.

TOMITA, T., KANEKO, A., MURAKAMI, M., and PAULTER, E.L. (1967). Spectral response curves of single cones in the carp. Vision Research 7: 519-531.

TYLER, J.E. and PREISENDORFER, R.W. (1962). Transmission of energy within the sea. In The Sea, vol 1 (ed. M.N. Hill), pp. 397-451. Interscience.

VAN DE HULST, H.C. (1957). Light scattering by small particles. Dover Publications Inc. New York.

UNIVERSITY OF MICHIGAN ENGINEERING RESEARCH INSTITUTE (1951). Tables of light-scattering functions for spherical particles. Edward Brothers Inc. Ann Arbor, Michigan.

WALD, G. (1939). On the distributions of vitamin A1 and A2. Journal of General Physiology 22: 775-794.

WATERMAN, T.H. (1954). Polarization patterns in submarine illumination. Science. 120: 927-932.

WATERMAN, T.H. (1955). Polarization of scattered sunlight in deep water. Deep Sea Research. Supplement 3: 426-434.

WATERMAN, T.H. (1956). Polarized light and plankton navigation. Pers. in Mar. Biol. (Scripps Institution of Oceanography Symposium, ed. A.A. Buzzati-Traverso). p. 429-450.

WATERMAN, T.H. and FORWARD, R.B. (1972). Field demonstration of polarotaxis in the fish Zenarchopterus. Journal of Experimental

Zoology180: 33-54.

WEHNER, R. (1989). Neurobiology of polarization vision. Trends in Neuroscience 12: 353-359.

WEHNER, R. (1983). The perception of polarised light. In The Society for Experimental Biology Symposium XXXVI; The Biology of Photoreception. (Edited by D.J. Cosens and D. Vince-Price). Society of Experimental Biology. pp. 331-369.

WELLINGTON, W.G. (1955). Solar heat and plane of polarized light versus the light compass reaction in the orientation of insects on the ground. Annuals of the Entomology Society of America 48: 67-76.

WESTERBERG, H. (1982). Ultrasonic tracking of Atlantic salmon (Salmo salar L.): swimming depth and temperature stratification. Institute of Freshwater Research. Drottningholm Rep. 60: 102-120.

WETZEL, R.G. (1975). Limnology. W.B. Saunders Co. Philadelphia.

WICKRAMASINGHE, N.C. (1973). Light scattering functions for small particles, with applications in Astronomy. Adam Hilger Ltd, London, England. 506 pp.

WING, B.L. (1977). Salmon food observations, p. 20-27. In: Southeast Alaska troll log book program: 1976 scientific report. Alaska Sea Grant Report 77-11.

YAGER, D. (1967). Behavioral measures and theoretical analysis of spectral sensitivity and spectral saturation in the goldfish, Carassius auratus. Vision Research 7: 707-727.

YAGER, D. (1969). Behavioral measures of spectral sensitivity in the goldfish following chromatic adaptation. Vision Research 9: 179-186

ZANEVELD, J.R.V., ROACH, D.M. and PAK, H. (1974). The determination of the index of refraction of oceanic particulates. Journal of Geophysics Research 79: 4091-4095.

VITA

Surname: NOVALES FLAMARIQUE Given Names: IÑIGO

Place of Birth: Bilbao (Spain) Date of Birth: 18 August 1966

Educational Institutions Attended:

University of Victoria	1991 to 1992
McGill University	1985 to 1990

Degrees Awarded:

B.Sc. (Biology)	McGill University	1988-90
B.Sc. (Physics)	McGill University	1985-88

Honours and Awards:

Fonds pour la Recherche et Aide aux Chercheurs Scholarship	1991-92
University of Victoria Graduate Teaching Fellowship	1992-93
Grant In Aid of Research (Sigma Xi Scientific Society)	1992
Clemens/Rigler Travel Grant	1991
University of Victoria Graduate Student Travel Grant	1991-92
James McConnell Entrance Award (McGill University)	1985-86

Publications:

Novales Flamarique, I., Hendry, A. and Hawryshyn, C.W. (1992). The photic environment of a salmonid nursery lake. Journal of Experimental Biology 169: 121-141.

Novales Flamarique, I., Griesbach, S., Parent, M., Cattaneo, A., and Peters, R.H. (1992). Fish foraging behaviour changes the

chlorophyll-nutrient relation in laboratory microcosms. Limnology and Oceanography (In Press).

Novales Flamarique, I. and Hawryshyn, C.W. (1992). Spectral characteristics of salmonid migratory routes from southern Vancouver Island. (British Columbia). Canadian Journal of Fisheries and Aquatic Sciences. (In Press)

PARTIAL COPYRIGHT LICENSE

I hereby grant the right to lend my thesis to users of the University of Victoria Library, and to make single copies only for such users or in response to a request from the Library of any other university, or similar institution, on its behalf or for one of its users. I further agree that permission for extensive copying of this thesis for scholarly purposes may be granted by me or a member of the University designated by me. It is understood that copying or publication of this thesis for financial gain shall not be allowed without my written permission.

Title of Thesis: Ultraviolet light in the salmonid environment

Author: [redacted]

Iñigo NOVALES FLAMARIQUE

27th April 1993