

Tributes and Reminiscences

To Celebrate the Life of

Shinya Inoué

October 26, 2019

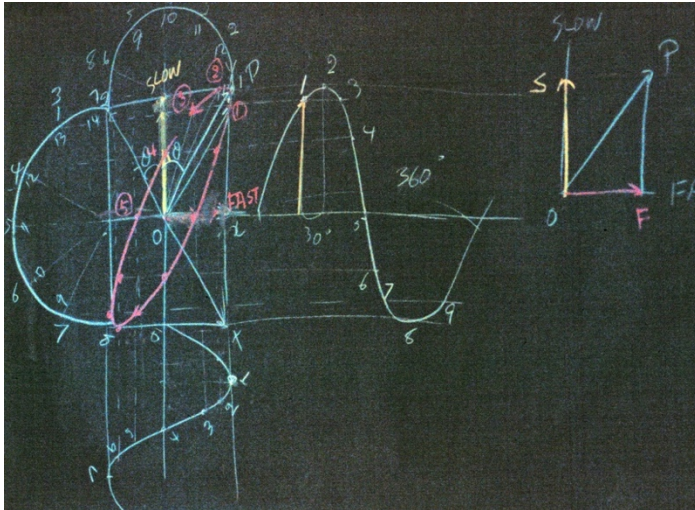
Marine Biological Laboratory
Woods Hole, Mass.

Collected by Rudolf Oldenbourg and Tomomi Tani

Jan Hinsch

A Moment of Grace

I drove to Woods Hole on December 3, 1978 to take part in the second annual "Optical Microscopy and Photomicrography in the Biomedical Sciences" course at the MBL. The course curriculum was conceived and inaugurated the previous year by Dr. Robert Day Allen and as a representative of the E. Leitz company I spent the following week assisting the students in the use of the company's instruments.



There were 16 students altogether and three instructors, Robert Allen, Nina Allen and Shinya Inoué. Soon the course had more applicants than places. In time two courses were held each year and the number of students doubled in both.

By comparison, the atmosphere in 1978 was more of a family affair, almost intimate. On Monday evening for example, Robert Allen would play his cello to welcome his students with 40 minutes of string music,

accompanied by Ernst Keller of Carl Zeiss who too was a dedicated cello player.

Two days into the course, Wednesday, it was Shinya's turn: Two morning lectures on the nature of polarized light. Shinya has the ability to turn a blackboard and a piece of chalk into a most powerful teaching tool. While he fills the chalkboard with the traces of the action of the slow and the fast ray in three colors Robert Allen walks into the auditorium, looks at Shinya's beautiful hopscotch and exclaims: "Shinya, t h i s is art!"

Shinya turns around: "You do it on the Cello".

Thoru Pederson

I can only say what many others would and will: Shinya displayed uncommon persistence as regards the reality of microtubules, against the slings and arrows of many opponents of lesser talent in optical microscopy, and his prescient development of video microscopy, also not without opponents, stands as a true milestone in cell biology. He will also be remembered, by those whose memories go back far enough, for his conscientious stand against certain academic deviants who had forgotten that the U.S. Constitution grants its citizens Freedom of Speech. These are the achievements and qualities for which we shall always remember him.

Susan Gerbi

I first met Shinya Inoué when I was a student in the MBL Physiology course in the summer of 1966, having just completed my first year of graduate school at Yale. Shinya's enthusiasm and scientific curiosity were contagious as he showed us spindles from unfertilized oocytes from the "ice cream cone" worm (the marine annelid *Pectinaria gouldi*) using a polarization microscope that he helped to develop. His lecture to the class about polarization microscopy had run overtime, so we took a brief recess for dinner and continued the lecture and labwork after dinner. Although it was approaching 11 PM, and the students were beginning to get tired, Shinya was still going strong, buoyed by the beauty of biology and mysteries of chromosome movement on spindles. His winsome smile, modest nature and wealth of knowledge of live cell imaging made him very approachable and much sought after by students and colleagues alike.

Decades later when I visited MBL for a collaborative project with Ted Salmon and Rudolf Oldenbourg, Shinya served as a valuable advisor. A few years ago, I had the pleasure of bringing my students from Brown to a festschrift at MBL for Shinya (July 14, 2011) that was aptly named "Listening to Life Through the Microscope". Despite his increasing frailty, Shinya's voice was strong and mind was sharp as ever, and he enchanted everyone in the audience with his talk. The cell biology community has lost a giant in the field as we wish Shinya good-bye.

George Langford

I can only say how lucky I was to have had Shinya Inoué as my mentor and to have worked with him during one of the most productive periods of his life. He taught me many life lessons but two stand out. Shinya would always say "find the best biological specimen to answer the question of interest". If the question is how chromosomes move to the spindle poles or how flagella bend or how vesicles move, find the appropriate cell type, i.e. use the wisdom of nature to help solve the problem. The second lesson was, "when imaging the cell, faithfully capture the information but also the beauty of the natural world". Shinya's aesthetic sense and his ability to decode the complexity of living cells will always be with me as part of his legacy. I remain eternally grateful to him for sharing his insights into the beautiful world of the cell.

Raymond E. Stephens

Shinya was truly remarkable in terms of the depth and breadth of his inquisitive mind -- in many ways, prescience personified.

Nina Stromgren Allen

I knew Shinya Inoué for 50 years and knew of him even longer as I had read some of his papers while in graduate school. I still remember how impressed I was by his beautiful movies using polarized light microscopy to show microtubules in living cells as presented at a talk at the MBL my first summer there. I really got to know Shinya in 1973. That year Bob Allen and I had a sabbatical at the MBL and Shinya had recently moved there permanently. Lansing Taylor and I were postdocs in Bob's lab and Ted Salmon in the Inoué lab that year. We would often eat lunch together at what was then an unassuming PJs, but now is Water Street Kitchen. There were wonderful discussions about making better microscopes and improve imaging in many ways as well as what and how to study and image living cells and cell motility in all its forms. I learned so much from Shinya in these discussions and visits to his lab and seeing his wonderful microscope at work. Shinya would share his knowledge and what a lot he had to share. I also learned that Shinya was a very kind and caring person.

Bob and Shinya were good friends as well as keen competitors and it was always fun to watch their interactions. Shinya had been at Princeton and later Dartmouth before Bob was at Princeton and later Dartmouth. Their careers were parallel in so many ways. They both developed new microscopic methods. The aim of both was to use these better imaging methods to see and record movements in and off living cells. And to instill in others the value of imaging living cells well. I think that they truly respected each other and were aware that advancing scientific knowledge was and should be an end goal for all scientists. On the whole as I recall it, that was the spirit of the MBL in the 1970s and 1980s.

One of the joys of teaching the Allen MBL Optics course at the MBL was the lecture we asked Shinya to present on polarized light and its uses in microscopy. These were elegant lectures with wonderful demonstrations and each time I would learn something new. Little did we know when those courses started how they would grow as Video Microscopy came on the scene and Light Microscopic imaging had a renaissance which is still ongoing. Later when I ran that course and started lecturing in it, Shinya would sit in the back of the lecture room and after my talk would gently and privately point out any errors I might have made. That was very helpful and kept me on my toes.

Shinya would also show the microscopy course students his laboratory and particularly his wonderful microscope in a darkened room. He would patiently and carefully answer their many questions. He was a fine teacher as was also demonstrated in his book called Video Microscopy. On a visit he saw my well-worn copy of that book and asked if he could show it to the publisher so that they would make the next printing sturdier.

After we in parallel invented video microscopy, Shinya had the fortune to have a son, Ted Inoué, who with his help developed Universal Imaging. Ted and his friend's knowledge of computers made it possible to develop relatively inexpensive and good imaging systems. Shinya was so happy and proud with that development and working with Ted was clearly so special for him. Shinya believed in family and with his wife Sylvia nurtured their family well.

It is an honor to have known someone so dedicated to Science, who was also a mentor to so many and a lovely man.

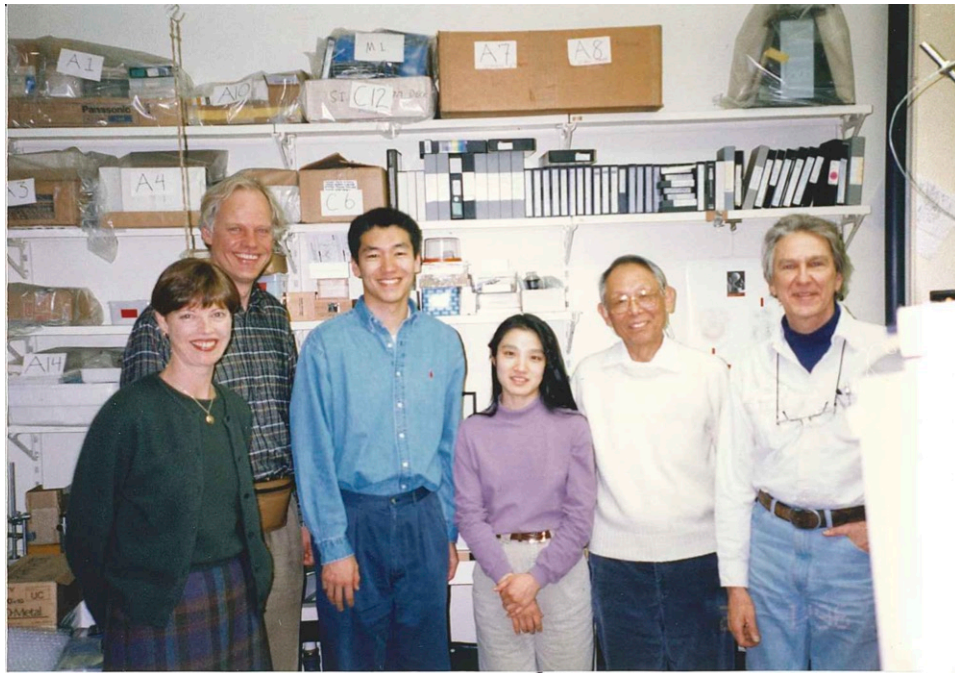
Tim Mitchison

Below are a couple of quotes I love from an essay co-written by Shinya Inoué and Ted Salmon (1995, *Mol. Biol. Cell*). These statements reflect the views of both co-authors, but somehow I hear Shinya's voice especially strongly. Listen to the living cell....

“...we should remember that nature sometimes reveals her most well-kept secrets through exaggerated displays found only in exotic cell types.”

“...we need, in addition to dissecting the molecules further, to listen ever more carefully to the living cell, and be prepared to be taught further unexpected paradigms.”

Rieko Arimoto



Shinya's research attracted people from diverse backgrounds; the lab was truly interdisciplinary, certainly steered my scientific interests into a new directions! From left, Jane MacNeil, Rudolf Oldenbourg, Keisuke Sukuzi, Rieko Arimoto, Shinya Inoue and Bob Knudson (1996), surrounded by piles of video tapes and neatly organized recording equipment.

Brian Salzberg

Shinya Inoué was, quite simply, one of the two or three most important microscopists of the twentieth century. His contributions, spanning well over half a century, transformed microscopy, and contributed enormously to cell biology. He was always accessible, and loved to talk about all aspects of optics, especially cutting-edge techniques. I was extremely proud to call him friend.

Rudi Rottenfusser

At the Zeiss US headquarters in NY, I attended many meetings when Phil Presley, the Zeiss Rep for the MBL in Woods Hole, kept on mentioning a famous Dr. Inoué who was considered a living legend by him and others.

I had no idea then, that I would be designated to become Phil's successor in 1995, after he unexpectedly passed away in the fall of '94. During the ensuing 15+ years, I finally had a chance to meet and experience the famous Dr. Inoué, both in his lab and listening in on many of his lectures. Typical for his well-executed presentations, he would use a computer, an overhead projector, and a white board interchangeably, moving constantly from one medium to the other. All of this to bring complex theories in polarized light across in an understandable manner! And everything brilliantly coordinated and synchronized!

Shinya was exceptionally focused on his scientific objectives, and he was undeterred by companies trying to win his favors to use his name for their commercial gain. A testament for this is the famous "Shinya Scope" which consists of components from all 4 major microscope manufacturers.

Despite Shinya's thorough knowledge and reputation in many fields of microscopy, he was refreshingly unpretentious and always willing to seek and accept advice from others. His humility did not diminish his status but rather lifted it up. He was not a very large man in size, but his spirit was large, indeed.

Thank you, Shinya, for having been an inspiration and a friend. I know that your legacy will live on as long as microscopy lives on.

Ron Vale

Shinya Inoué had a big influence on my life. I remember meeting Shinya when I was a graduate student at the MBL in Tom Reese's lab in 1983-1986. Working on microscopy myself, Shinya was a heroic figure who pioneered live-cell imaging, built magnificent microscopes and led the digital era of microscopy. Shinya was filled with a passion for science that burned strong throughout his life. However, I also remember him as a kind individual who always had a twinkle in his eye. We would have nice talks in the dead of winter in Woods Hole; he looked after me then and helped me in my career. At the close of my time in Woods Hole, Shinya suggested and effectively arranged for me to do a "Dan Fellowship" in Japan. Working in Kyoto, Japan at age 27 proved to be a very influential 3 months in my life, and Shinya's introduction seeded my long-term connection with many Japanese scientists.

My wife Karen Dell and I later wrote an article in the *Journal of Cell Biology* in 2004 in honor of Shinya's receiving the 2003 International Prize in Biology and a special meeting held in his honor in Japan. The article concluded by saying:

"At 83-years young, Inoué, with his love of science, his continued keen insight into cell biology, and his unwavering support of young scientists, continues to teach us a great deal about biology, microscopy, and how to conduct ourselves in the pursuit of scientific discovery. The 2003 International Prize for Biology could not have been bestowed on a nobler scientist and individual."

Remarkably and thankfully, Shinya continued to inspire us for 15 more years after this article was written. He was indeed as noble a scientist and individual as I have met in the scientific profession.

Stephen Smith

Shinya Inoué was a researcher of singular importance, opening new fields of cell biology and new approaches that truly and profoundly changed the ways we think about and study living cells. His contributions have withstood the test of time in a rapidly changing world of biological science to an extent that is truly extraordinary. That alone would certainly be enough to demand accolades like “great” and “towering giant,” but Shinya was much, much more.

Shinya set a high standard for sharing one’s wealth of knowledge and bench savvy that is not only unmatched but has propagated to our entire field, setting a tone of cooperation that I believe has been a key factor in the stunning successes of modern cell biology. Here I’ll note just his extremely influential efforts in the teaching and dissemination of quantitative microscopy methods. His courses at the MBL not only spread the fundamentals to generations of students (and professors) but also established an extraordinary model for dialogue between academia and industry. The force of these courses has also been multiplied many times over by the “imitators” they have spawned across the country and around the world. Again, this alone would be enough.

Shinya was something else still, however, perhaps even more precious. His personal kindness and generosity were also unmatched. I know because I was a beneficiary and so were many of my friends and their friends. I can think of no one else who is held in such high regard for such traits. I believe that part of Shinya’s legacy is having inspired all of us to treat each other a little better as we go about our work. He will be sorely missed but fondly remembered for as long as we live.

Stephen Ross

I recall how generous Shinya was with his time to mentor and teach people. Specifically, when one of my great mentors, Katsuji Rikukawa (who was a friend of Shinya's, and designer of Rectified Optics for the 4DLM, Shinyascope), came to visit, Shinya would always ask him and Matsui-san (another of our Optical Designers, who designed Nikon's DIC systems) optical "Test Questions," to challenge and teach them. Also, for some time, in that tradition, he would ask me optical questions, usually with a warm smile on his face, just to test my knowledge, when I would stop by to visit.

Amy Gladfelter

I will never forget working on an R01 application with Tomomi Tani and Rudolf Oldenbourg with help from Shinya. His insightful, critical and energetic interjections and discussions over email happened generally in the wee hours of the night -when he was about 90 years old! Shinya's drive to help us improve the science and how we communicated it continue to inspire me to be generous with ideas, time and care for the development of others.

Larry Cohen

Shinya was my teacher in the 1962 Physiology course. I became an admirer of his.

Then in 1966 I went off to England as a postdoctoral fellow with Richard Keynes. Twenty years earlier Richard and David Hill had measured light scattering signals from crab nerves during trains of action potentials. A new tool, the Computer of Average Transients, became available and Richard had me continue optical measurements with the higher time resolution and sensitivity that the new tool made possible. Eventually we were able to record birefringence signals during the action potential of the squid giant axon. Unbeknownst to us, Bob Barlow and Shinya were trying to do the same experiment at the MBL. When I returned to the MBL in 1969, we discovered the overlap. Both Shinya and Bob supported me then and treated me with kindness for the rest of their lives.

In the email that I received from Shinya's son Ted, there was the following sentence:
"I know you were important to my father - he had only kind words to say about you, both as a scientist and as a friend."
"

I was so touched. And also filled with regret. That I had not shown Shinya my affection and respect often enough and openly enough.

In my Conservative Jewish tradition it is said: "Those who have died live on in the hearts and minds of the people who loved and cherished them." So it is with Shinya.

Keigi Fujiwara

Although I was a graduate student in the Biophysical Cytology Program at Penn for 5 years and took 2 courses offered by Shinya, I do not remember having any personal or long conversations with him during that time except for on one occasion, which I will describe in a minute. This was partly because Shinya was not my direct thesis advisor, but the main reason was that for me, Dr. Inoué was so far above me that I didn't know how to approach him, let alone what to say. Throughout my Penn years, I could never call him by his first name; he was Dr. Inoué. Period.

So, the only time I did talk to him was when I wanted to take the Physiology Course at MBL. I went to his office and asked him if he had time to speak with me. He looked surprised, but pointed to an empty chair. I sat and clearly remember feeling very awkward, but I must have begun talking. He first listened and then asked me a few questions. I was so surprised that he was truly interested in what I had to say and responded to me with smiles, softness, and support. He then told me that the deadline for application had long passed. Hearing that, I was ready to thank him and leave, but he told me to stay then picked up the phone. I remember Shinya saying on the phone something about the uncertainty of the course being offered the following year. Before ending the call, they discussed matters that had nothing to do with me. After the call, Shinya said, "I think you're in." I thanked him of course, but I thought what a powerful person he was. I was of course very happy to hear this, but I remember thinking "I think you're in" is not a definitive statement. However, I now know that when Shinya says, "I think...", that means IT IS SO.

It was after I left Penn that Dr. Inoué became Shinya. Although no paper was published, we did many experiments together at MBL, mostly him microinjecting antibodies into fertilized sea urchin eggs. Through these experiments, I got to know Shinya well, his true fascination about biology, uncompromising technical perfection, and meticulous record-keeping. I remember that whenever I said something that he did not agree with, he would not say anything but would tilt his head sideways slightly and look at me with a smile. I have many fond memories of him: having a countless number of meals at Captain Kidd, dining at a restaurant in Kyoto with Shinya and Sylvia, taking long brisk walks together in Osaka, collecting sand dollars, catching grasshoppers, and chatting with him at the ASCB meetings. Like all of us, I will miss him and am better for having known him, a person who was a great teacher, incredibly creative scientist, and friend with warm smiles.

Michael Shribak

We met with Shinya in March 2000, after I came to the USA for the first time. After this visit I joined the Architectural Dynamics in Living Cells Program where Dr. Inoué was the Program Director. Since then we interacted closely on scientific and personal issues, our families became friends. Dr. Inoué let us to stay in his home and was our landlord for many years. He taught me biology and helped us to navigate through our new life in America.

I would like to mention Shinya's spirit of innovation. Shinya always used the best technologies, which were available at that time. If a necessary technique did not exist then Dr. Inoué invented it. This list includes polarization rectifier, centrifuge polarization microscope, polarization fluorescence, local fluorescence illumination, etc. Shinya's contribution to science is impossible to overstate.

Shinya inspired many people for new inventions and discoveries. He inspired me on the invention of the quantitative orientation-independent microscopy, polychromatic polarization microscopy, improvements of polarization rectifier, developing mathematical model of polarization fluorescence in GFP, etc. We continued to meet with Shinya regularly even after his retirement in order to discuss new microscopy developments and Shinya always had comments and gave good advice.

Shinya is one of the most remarkable people with whom I met in my life. This is big loss for me personally and to my family. Shinya is my role model. His memory will live with me forever.

Louis Kerr

I came to the MBL in 1980 and very quickly met Dr. Inoué through my duties of assisting in the MBL short courses, which included the AQLM course.

The other day while on the water I saw a sun dog and it reminded me of Shinya as he was the one who introduced me to these phenomena. The sun dog is caused by the refraction of sunlight by ice crystals in the atmosphere. Seeing the sun dog lead me to think about the time he told us that we humans can perceive polarization. An example he gave was Haidinger's brush and he said it could be seen as a bluish bow tie while looking at a clear blue sky early in the morning. A few days after his presentation I came to work and the sky was blue so I went outside the Lillie building and laid upon the concrete pad and looked up. After a few minutes I could see it! I told Shinya about it and I think he was surprised that I would try but impressed that I was able to see it.

On another occasion when his son Ted was in high school, he and Shinya asked me if I could coat a hand polished mirror with aluminum to use in a telescope they were building. They came to me because we had a vacuum coating device that could evaporate aluminum. I said sure thinking we would load some aluminum foil on a filament, put the mirror on the base of the instrument and do a vacuum evaporation run. Of course, I wound up getting a lesson about how the coating thickness is based on the amount of aluminum and the distance from the aluminum to the mirror. Shinya and Ted knew the thickness they wanted so we were able to calculate the other two variables. It worked out well!

I was impressed with Shinya's interest in civil engagement and trying to make a positive impact while acting with humility and also being prepared. Over the years on many occasions in meetings and lectures in the Lillie Auditorium I was witness to this effort. If you go into the Auditorium through the back doors Shinya liked to sit about halfway up in the left side section. I am sure he had a good reason to select this area, but I don't know why?! Regularly during MBL Society meetings, general institutional meetings and lectures Shinya would raise his hand to deliver a well thought out question, comment or suggestion. He presented these without exaggeration and was always well intentioned.

A comment about his ability as a teacher. I really enjoyed listening to Shinya lecture because his excitement and attention to detail was visible. He knew his subject and presented it in interesting ways. He liked to draw detailed diagrams on the board, use the overhead projector and use props, such as two pieces of the same type of wood cut in orthogonal directions and then using a tuning hammer he would demonstrate how they made different tones. This represented the two different fields of polarization. I was able to work with Shinya to produce the muscle-thin section slides that very eloquently show the utility of polarized light microscopy. Shinya left nothing to chance, the sample – striated frog leg muscle in the tensed state, fixation protocol, embedment, section thickness, orientation on the coverslip, mounting media, etc. All these things were thoroughly thought out. (*Continued ...*)

(Louis Kerr, cont.) In the end, Shinya was a great mentor to me without him even trying to be. He was humble, detail orientated, quietly humorous, respectful and wise.



Shinya and Louie Kerr in AQLM class photo, year unknown.

Joseph and Jean Sanger

On behalf of Jean and myself, I would like to say a few sentences about Shinya.

It was a different era in the 60's (Dartmouth Medical School, and then Penn) and 70's (Penn), so we do not have any photographs of Shinya.

The attached photo is of Jean and me on the first full day of our honeymoon in Bermuda, September 13, 1964. We were married in Falmouth on September 12, 1964, after the Physiology course was finished. Shinya and Andrew Szent-Gyorgyi attended our wedding reception.

Shinya arranged for us to be his two graduate assistants in the 1964 Physiology Course so that we could spend the summer of 1964 in Woods Hole. Jean grew up in Falmouth, and her family has had a long connection to WHOI.

Shinya was not only our scientific hero, but he looked out for students in his Department of Cytology at Dartmouth. My comments would relate to his kindness towards Jean and me at Dartmouth, but also at Penn. Jean and I were in the Anatomy Department at Penn, and Shinya was in the Biology Department at Penn, only a very short trip between the two Departments.



Paul Forman

I learned one of my most important lessons in life from Shina Inoué when I was working as his research assistant. I assisted him with a polarizing microscope he was developing, and once complained about the awkwardness of the focus mechanism. Two days later he handed me a gadget that completely eliminated the cause of my complaints. It was a sophisticated, not-at-all-obvious solution, and I asked how he came up with that solution. He explained that he simply believed there must be a good solution and worked at it until he found it. “I don’t understand,” I replied.

He asked me “Can you balance a raw egg on end?” “Sure. Just sprinkle salt on the counter and the egg can be balanced,” I replied. “No, no. No tricks. A raw egg—balanced on a steady tabletop.” “No, I do not think it can be done. I’ve tried it before without success,” I replied.

He took a deep breath, looked me in the eye, and asked whether I trusted him. “Of course.” “Well, then trust me when I tell you it is quite possible to balance a raw egg on end, no tricks. Now, go to the biology refrigerator down the hall, get an egg, bring it back here, and try it,” he demanded.

It took me less than a minute of trying to balance that egg, because I had perfect confidence it could be done. That lesson has guided me so many times in my life, in so many important ways.

Lesson learned: If you believe something can be done and it is worth doing, be persistent and just do it.

Brad Amos

I first started communicating with Shinya in the 1970s, when I had completed my PhD on the contraction of the spasmoneme, the contractile organelle in vorticellid ciliates, which underwent a dramatic decrease in birefringence during contraction. I knew of him, not only from his exemplary published work on the mitotic spindle, but also through his ex-student Art Forer, whom I knew well in Cambridge at that time. I regarded Shinya as an august figure many years my senior and regret that I never became close enough to him to have any personal anecdotes.

I have had many contacts with Japan and once asked Shinya if he had a Japanese soul. He became very thoughtful but would not answer. My own theory is that, as the son of a Japanese diplomat and early companion of the Japanese royal family he was Japanese in his reserved public persona but enthusiastically adopted competitive and inventive Yankee values, which perhaps caused an internal conflict.

Shinya might have been amused to know that I had the honor of demonstrating, in the 90s, our prototype confocal microscope to Crown Prince Fumihito (brother of the current Japanese Emperor), who immediately took control of the microscope focus and asked, 'Do you use special fixatives for the confocal microscope or just conventional LM fixatives?' He astonished our research students at LMB Cambridge by knowing all about restriction enzymes and discussing his research on the catfish in the Imperial Palace gardens.

In 2003 my association with Woods Hole ended and I set up in Plymouth UK a microscopy course initially modelled on the AQLM course. I have borrowed some of Shinya's and Ted Salmon's ideas for lecture demonstrations and the course has run annually every year since then, with some familiar US lecturers, including the now-deceased Dick Haughland and Roger Tsien and attracting the best students from Europe and beyond.

My wife, Linda Amos, is now retired from the MRC Lab of Molecular Biology, as I am. Due to my wife's health, I cannot travel from Cambridge easily, even for short trips. Please convey my apologies and greetings to friends from the US scientific community, whose company I used to enjoy so much in Woods Hole, including Joel Rosenbaum, who very kindly allowed me to attend the Physiology Course as a student, which allowed me to learn a great deal that had been missing in my haphazard education.

Mai Tran

I was a former PhD student and postdoc in Rudolf Oldenbourg's lab from 2012 to 2019. When I joined Rudolf's lab, Shinya had not visited the lab often. However, I was so lucky to use Shinya's office. Here, I can learn from how he organized his stuff, especially his books which are invaluable. Sometimes, I can see his helpful notes while reading the books. In addition, his old work was still valuable for my work on microscopy. Actually, I found the ideas based on his work.

I believe that Shinya's work on cell biology and microscopy will be immortal. I always consider him my indirect mentor. I don't know him much in person but from my respected colleagues and people in Woods Hole, I know Shinya is an amazing and great person. I really admire him and his love for science.

In summer 2012, our lab was suffering considerable financial difficulty. Shinya was very generous to use his own money to support us. We are really grateful for his support not only financially but also emotionally.

I remember when I was first at MBL, someone asked me "Do you know Shinya? Have you seen his microscope?". Now, it's my turn to ask a new MBL visitor if I have a chance. "Do you know Shinya Inoue? Have you seen his microscope?"

I would like to express my deep gratitude to him. Rest in peace. Shinya will be missed by us, forever.

Hideo Hiruma

Our relationship started when Shinya gave us some claim on our camera for microscope. The trouble was a simple one. It was fixed in five minutes. But it happened simultaneously that so called "real time subtraction on the image" which named later on as VEC (Video Enhanced Contrast) was invented. We continued collaboration with him to develop high sensitive laser scanning microscope. This technology is utilized in semiconductor failure analysis field effectively.

I remember many time of driving to the MBL from Boston and spend a wonderful time with Shinya and hope for a peaceful heaven for him.

Hiro Ishii

When I first started to work at the MBL as a postdoc, Shinya-sensei kindly came to his lab and taught me how to use his microscope. It was the strict winter season of 2015. I was truly impressed with his passion and sincere attitude toward science. Shinya-sensei was my dream scientist who taught us how the living cell is beautiful and unique and the true worth to study the biophysical mechanisms deeply as a scientist. May he rest in peace.

Issei Mabuchi

I first met Shinya-san when I went to the U. of Penn as a postdoc in Lew Tilney lab in 1977. I was lucky to collaborate with Shinya-san and his graduate student Dan Kiehart. In every summer of the past 15 years or so, I used to visit his office or house and enjoyed talking with him. His passion for science inspired me and his excellent memory surprised me every time.

Kaoru Katoh

It was with great sadness that we learned about Shinya's passing. Dr. Tani was kind enough to e-mail us the sad news.

During my postdoctoral period, I learned many things from Shinya. I still remember Shinya had a lecture on polarized light microscopy in a seminar room in MBL. He cut down a big and special calcite crystal to show us an optical axis of the calcite. The course of lectures on polarized light were held twelve days! I was so impressed by the lectures.

Shinya did paper works in the morning and did experiments in the afternoon. During his experiments, Shinya sometimes said to himself in Japanese words "I should be scolded by Dr. Dan". I heard the short monologue a couple of times. Shinya might think back to his young days with Katsuma Dan.

After I left Wood Hole, I saw Shinya a couple of times in Japan. Last time I saw Shinya in Tokyo in 2010, Shinya's scientific presentation was exciting and he looked fine, but he told me this might be his last visit to Japan because of his age.

I learned that a Celebration of Shinya Inoué's Life will be held at the MBL on October 26, 2019. Although we are in Japan far from Woods Hole, we would like to do anything what we can do and share memory of Shinya. There are many collaborators of Shinya in universities and companies in Japan.

Shinya was the greatest scientist for us. We still remember his voice and performance in our heart.

Kazuhiro Oiwa

When I was an undergraduate student of the Department of Zoology, the Tokyo University, I spent a week in the Misaki Marin Biological Station (MMBS) and met Katsuma Dan. Although he had been already retired from the university, he spent many days in MMBS for his research. He kindly showed us the cleavage of the eggs of *Clypeaster japonicus* with an optical microscope. At that time, I happened to hear the name of Shinya Inoué from Katsuma Dan.

When I became a graduate student, the book entitled “Video Microscopy”, which Shinya Inoué authored, was one of the most influential books on me. I have still consulted this book for my present research. As a postdoc, I received his kind words on our research achievement in 1990 on the centrifuge microscopy applied to *Nitella* actin cables and these words very much encouraged our group and me. I was thus inspired by his wisdom, his critical thinking, and scientific objectiveness.

The photo attached reminds me of the scene that Shinya Inoué had a lecture on the wood stage of the Noh theatre in Nara Kasugano International Forum, where the symposium was held honoring Inoué’s 2003 International Prize for Biology award and his pioneering spirit in light microscopy. It was a great honor and privilege for me to have a chance to give a talk in the symposium.

Shinya Inoué will be greatly missed by his family, his students, his colleagues, and his friends.

Kazuhiro Maeshima

I am so sorry to hear the sad news about Shinya-sensei. How hard it is to lose him whom the MBL and people in the field, including me, have respected so much!

It was in June 2010 when I first visited the MBL Cellular Dynamics group and met Shinya-sensei. After giving a seminar on our chromosome organization work, I talked with him. I was a bit nervous because he was a legend of live-cell imaging. I remember that during our discussion he mentioned a very old story on a chromosome peripheral structure in 1930s-40s. To be honest, I could not understand well what he meant at that time. However, much later, I realized his point was very important and was so much impressed by his deep insight into cellular organization in living cells. Since my first visit in 2010, I sometimes visited MBL and had exciting time. Although I have not had an opportunity to meet Shinya-sensei, thanks to his support behind us, our collaboration on OI-DIC imaging with Michael Shribak and Tomomi Tani has been going pretty well [Imai et al. (2017) Mol Biol Cell. 28, 3349-3359]. We are also planning to see birefringence in the chromosome. It is really sad that I cannot discuss our fruitful outputs and interesting plan with Shinya-sensei anymore.

Mahito Kikumoto

We, Prof. Oosawa, his wife and me arrived in WoodsHole at 1993. At that time, Shinya's group had the members of, Rudolf Oldenbourg the scientist and his assistant Guan May, Andreas Stemmer the Post-doc. from Swiss, who is constructing bio-AFM, Jane the secretary, Barbara the assistant of Shinya.

Here are epsodes related to Shinya during my stay and etc.

[Ep.1] Prof. Fumio Oosawa was invited to the summer courses in Woods Hole in 1993 and stayed first time there. He had lectures in Microscopy course directed by Shinya in May, and in Physiology course by prof. Tom Pollard from July to August. He brought me to assist his lectures. I heard firstly my stay will be 2 weeks and a few days. After 2 weeks, I sad how nice place WoodsHole is! And Oosawa-san said to me, do you keep to stay until next his visit in July for physiology course lecture?. It was decided that I would be extended my stay and make some experiments without my recognition in somehow. Shinya or Oosawa-san should pay my staying fee in somehow. Because it was the issues between Oosawa-san and Shinya, I was grad-student and could not recognize those situations in real. Anyway, Shinya and Oosawa-san should support my stay in Woods Hole.

[Ep.2] During stay, I broke temple of my glasses frame with putting on the bed carelessly. Shinya helped me to ask his optimist to repair my frame in makeshift way. I got back my eye-sight soon. After that, I ordered parts of that from Japan.

[Ep.3] The airline ticket for return to Japan was discard because it was discount ticket when my stay was decided to extend. I had to get reservation to return to Japan at airline corporation reservation. However, August was busy season, I could not get ticket reservation to return even before the limit of stay without visa (90 days). 1 week before limit day, I was at a loss and talked Shinya this situation and ask him to get ticket. He could help me to get reservation and ticket for my return in somehow. Therefore, I could return to Japan for 88-days without illegal stay.

[Ep.4] Shinya also recommend me to talk with Shimomura-san in downstairs (1F). I often met him, because Shinya's lab was 3F and Prof. Shimomura's lab was 1F in same stair. I already knew that he found aequorin and GFP from the jellyfish, the aequorin was used for the first observation of calcium-wave in the fertilization of Medaka egg. I visited his lab and talked with him about mainly aequorin, not so much GFP. That was nice time to know his personality. He was purely the scientist as same as Shinya.

[Ep.5] In the experiment of the effect of electric field for actin polymerization during stay, because I made experiment from the setup of experimental condition and measurement system, I often went around here and there to find wanting but lacking instruments or materials. In the meantime, someone helped me to get instruments or (*Kikumoto, cont.*)

materials from somewhere. Of course he helped me directly, but it also might be Shinya's deep considerations to support me.

[Ep.6] It was in the Independence Day at night, Shinya invited me and Universal Imaging's programmer, Pete Peterson, to his home to see the fireworks display in the Falmouth's beach for relaxation. We could enjoy fireworks and stay in his home and spent elegant and full time. It was strong impression that we heard the sound of the trash can scavenged by raccoon. It was excellent break time.

[Ep.7] When Shinya and Bob worked for automating process combined focusing between condenser and objective lenses step by steps, the accident of motorized focusing system was happened. The runaway of motorized system was rose up and crashed the stage and objective lens. Shinya calmly turned off the motor power supply to avoid secondary accident. Next week, Gordon Ellis came to repair the rotational stage. I learned from this accident that we have to keep cool even when the accident occurred.

[Ep.8] Shinya tried to observe the bacteria flagella when he knew Dr. S.M. Block observed bacteria flagella with DIC microscope. But that day, he could not observe bacteria flagella and was chagrined strongly as "Why I cannot observe flagella? Something strange." He was eager to observe flagella by himself. I felt it's the passion of researcher and he still have it. I learned the strong passion needs to research.

[Ep.9] Here is microscopy division of some institute in Nagoya university, they have Shinya-scope but not worked. I found it and then rebooted 3 years ago. Firstly, it was presented as one of the monument same as ZEISS Axiomat and did not work, because it's a junk state. I felt it was no worthy of Shinya scope, so I decided to reboot. I searched parts to need and reconstituted to observe with DIC microscope without manual and parts list. I found many parts has lost, it was difficult to reboot to the original state as before. I could reboot as only low resolution DIC microscope but high contrast in somehow.

[Ep.10] When Shinya came to Japan, I always went to meet him in somewhere in Japan. He could remember me and addressed me as "young friend" to come to meet him at that day. It's an honor for me that he called me a friend.

During stay in Shinya's laboratory, it was very happy and valuable time because I could meet many peoples (Ted, Silvia, his previous students, Ray Stephens, Tam, Lui, L.Tilney, Ted Salmon, Mario Burgos, Shahid Khan, Takashi Shimizu, M.P.Sheetz, and so on) and to take summer-course lectures without fee. Those were realized caused by supporting by Shinya for my stay. I feel Shinya is one of my "Sensei" or a model of the researcher same as Oosawa-san.

God rest Shinya's soul.

Makoto Goda

It was the spring 21 years ago that I met Shinya for the first time. After that, he had taught me various microscopy methods and cell biology every summer. His kind teachings are now most important foundation of my present scientific works. And, I am very honored by that I had had conversations with Shinya about various biological subjects every summer as a good friend. It was most pleasure things for me in Woods Hole.

He will live on in my heart and mind as a great teacher of science and as a good friend, forever. I will definitely never forget his kindness.

Naoki Noda

This is my favorite story of Dr. Shinya Inoué and me. Sometime, I bring back these wonderful experiences in Woods Hole.

On Shinya's 89th birthday, January 5th, 2010, I presented him with a photograph of the famous jelly fish, *Aequorea* which I had taken. Shinya glanced at it and said, Thank you, but the best birthday present for me is when you make interesting discoveries and produce meaningful research results." Shinya's words were emblematic of his dedication to science and the passion with which he raised his younger colleagues. Those words have been with me ever since.

In the summer of 2010, I happened to discover that live cells, or lithocytes, moved along on the cilia to build the gravity sensing organ of the comb jelly. On Shinya's 90th birthday, January 5th, 2011, we discussed this finding and I enjoyed our discussion very much. I believe Shinya enjoyed that too.

At that time, I thought this discovery was a birthday present from me to him. But thinking back on it now, I realize this discovery was actually a present from him to me. Shinya has given me many new perspectives and really expanded my scientific field of view. Thank you so much, Shinya!

Mizuho Shimizu

It is exactly 20 years since I first met Shinya at MBL, when Yokogawa sent me to demonstrate the CSU-10, spinning disk confocal scanner, at the AQLM course. I used to be an industrial microbial researcher before joining Yokogawa but my knowledge and skills in microscopy was quite poor. Shinya very kindly and patiently gave me personal elementary lessons how to clean lens and very basic Koehler illumination method, which I still remember vividly. Ted Inoue helped me all the time to run the Argon laser and a video camera to enable running the CSU-10 and successfully demonstrated direct viewing of live confocal image, which resulted in cheers all the time. Shinya, himself was the very first researcher to find the potential of the CSU-10 long before it had been commercialized, as Takashi Akiyama writes.

Especially, Shinya had discovered significantly reduced laser photo-damages with the microlens-enhanced spinning disk confocal method far ahead of any researchers. Shinya's positive endorsement was most important for the acknowledgment by notable biology researchers such as Ted Salmon and Ron Vale, who saw the live confocal images with the prototype CSU-10 at Shinya's MBL laboratory and became one of the earliest CSU-10 customers. Exactly, "seeing is better than hearing".

In addition, since the CSU-10 is just a scanner, it is necessary to set up a full system consisting of a microscope, laser light source and a camera, for which Shinya's kindest advises based on his profound knowledge were immeasurable benefit. Without Shinya, the CSU-10, a product of quite a stranger in the microscopy society, could not have succeeded at all !

When Shinya was awarded the International Prize for Biology in 2003, the most prestigious science award in Japan, he so kindly included me into his invitation list for the prize-awarding ceremony and reception where the Emperor and Empress attended; the best memory in my life. The Emperor, a biology researcher himself, made a stirring celebration speech to introduce Shinya's great achievements in biology and microscopy quite exactly and in detail, expressing his sincere respect and understanding of Shinya's achievement that Ted Salmon and Clare Waterman, who sat next to me at the ceremony, were amazed by the intelligence of the Emperor in comparison with then US president. After the ceremony, a reception took place in a relatively cozy room, where the Emperor and Empress stood all through and talked with Shinya and all of his family members quite affectionately, including young grandchildren. Thanks to his invitation, I too had once-in-my life experience to personally talk with the Empress! Very unfortunately, no personal pictures were taken since cameras were strictly prohibited.

Looking back, Shinya was always open, kind and generous, never showed anger or frustration, had never looked down on industry people, and always eager to explore new technology all through his long life.

I believe his great achievement and grand character shall be remembered and respected for a long time.

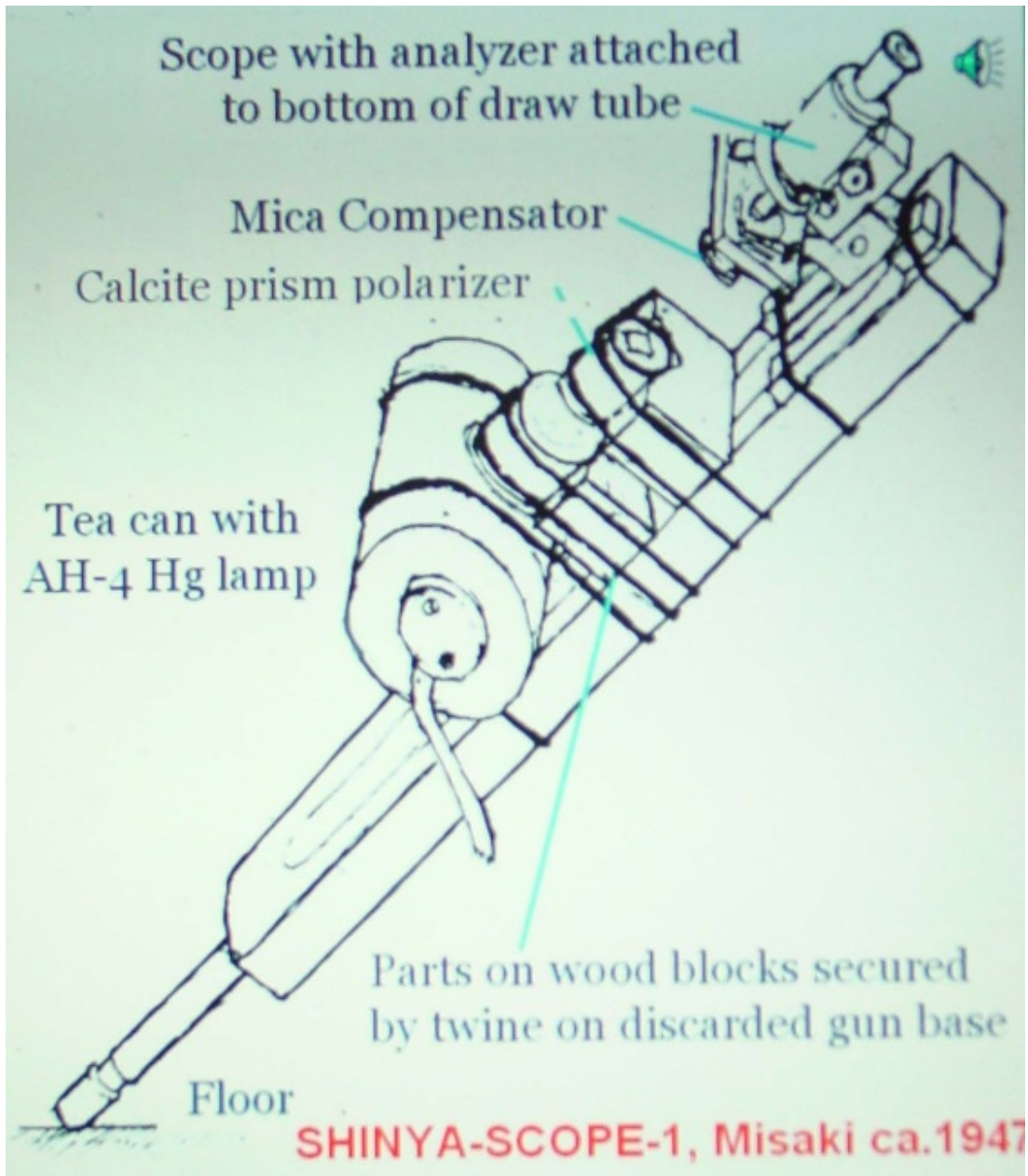
Motonori Hoshi

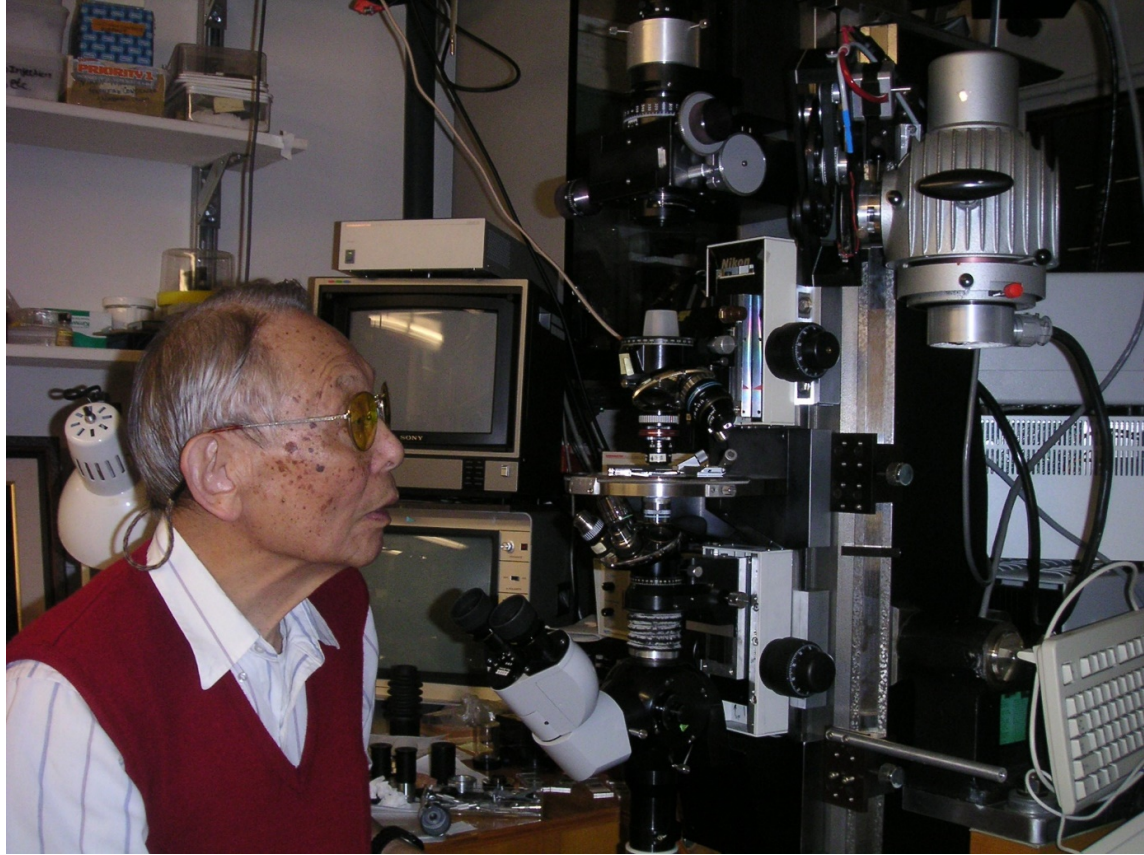
Inoué-san always welcomed me and enthusiastically explained me his new findings whenever I visited him at MBL. His never-fading passion for biology always encouraged me. I saw him time to time at MBL, but did so only twice in Japan, namely at the Presentation Ceremony for the 2003 International Prize for Biology¹ and at a symposium in Tokyo.

The research field for “the 2003 International Prize for Biology” was Cell Biology, and the Prize was awarded to Dr. Shinya Inoué for his outstanding contribution to the advancement of research in the field and in fundamental biology in general¹. When I was asked to explain his great achievements to the Emperor and Empress, I decided to concentrate my talk to his pioneering work on the analysis of dynamic processes in the living cells, such as cell division, by sophisticated light microscopy and asked him to send me some photos of him working with the famous Shinya-Scope. He sent me two photos below and some others.

The first photo is his drawing by memory of the first model of the Shinya-Scope that he made at Misaki Marine Biological Station, University of Tokyo in 1947 or so. As explained on the drawing, he had assembled some parts of scientific instruments together with a used tea-can probably from his kitchen and a discarded gun-base which might be not so difficult for him to find soon after the World War II. The second one shows Shinya working with a far more sophisticated Shinya-Scope shortly before his departure for the Ceremony held on December 1, 2003. These two photos clearly tell us his life as a great scientist.

By using the Shinya-Scope improved day after day, he gave us dynamic images of living processes in cells, and he opened, I believe, the heavy gate leading to ‘one-molecule observation’ and further into ‘one-molecule biology’ developed by much younger scientists such as Dr. Toshio Yanagida. (*Photos on following pages*).





Takashi Akiyama

All of us are grieved to hear of Dr. Shinya Inoue's passing and I would like to offer my heartfelt condolences to his family and friends. Please allow me to say a few words about Dr. Inoue's influence on Yokogawa.

The name of Yokogawa and our products have only recently become well known internationally. Amongst the products that have contributed to Yokogawa's reputation is the CSU (Confocal Scanner Unit), which enables the observation and imaging of living cells in real time. Not so long ago, Yokogawa was an unknown company at in this field, but a chance encounter with Dr. Inoue contributed to a major change in our position in confocal scanning technology.

In the middle of October, 1995, the late Dr. Harunori Ishikawa, the former president of the Bio Imaging Society of Japan, told us of his good friend in the U.S., the distinguished Biophysicist and Cell Biologist Dr. Shinya Inoue. He let us know that Dr. Inoue would soon be coming to Japan and that he might contact us. Soon afterwards, Dr. Inoue called us and suggested that he could make short stop by our facilities the morning of Saturday, November 25, as he planned to visit his sister who lived close by in Mitaka. At that time, we were not familiar with Dr. Inoue's work, but he came recommended by Dr. Ishikawa, so we were very happy to have him visit.

We do not normally conduct business on Saturday, but we welcomed him to Yokogawa around 10 a.m. Although he only intended to stay until noon, we were captivated by his ideas and he graciously remained with us until after sunset. As he departed that evening, he surprised us by saying: "Why not bring the CSU to MBL when it is completed?"

At that time, we had little confidence in our abilities to promote the CSU business, but over the next year, we took up his offer and, although we were not at all certain the work would be within our capabilities, we brought the completed CSU to MBL. Shinya graciously worked with us as we undertook a serious investigation of this technique. After few days of work and a demonstration of the instrumentation, Shinya kindly gave us his comments.

"Even though there are some limitations on performance in the CSU," he said. "I recommend that you release it into the market as it has unique features."

(Akiyama, cont.) Because of his inspiration and encouragement, we made the decision to begin the sale of the CSU through our international distribution system.

I am struck by the conjunction of events that changed the course of our company: if we had not known Dr. Ishikawa, if Yokogawa were not located in Mitaka, and if we had taken the Saturday off on that particular weekend in November, then Yokogawa would likely be quite a bit different today than what we have become. This combination of events and Dr. Inoue's guidance and encouragement have permitted us to become a leader in this field.

During our long relationship with him, we have benefitted greatly from his polite manners and kind attitude towards us, and we know that other companies in the Life Sciences industry share our high esteem.

Dr. Inoue's distinguished career in academia and his generous and kind personality have been recognized throughout the world. We at Yokogawa are deeply grateful for not only his contribution given to us when we were yet an unknown company, but also for his thoughtful guidance and encouragement.

Take Nagai

I would like to share a photograph taken at Friday Harbor in 2004 by my Sony camera. 2nd person from the left is Shinya-sensei. You can also find Roger Tsien, Martin Chalfie and Osamu Shimomura. A guy wearing striped shirt and sitting in the front-middle of this picture is me.

At this meeting, I asked to Shinya-sensei about how to compensate depolarization caused by light-passing through an objective lens with high numerical aperture. At that time, I wanted to use fluorescence polarization to detect homo-FRET because I found that a FRET-based calcium indicator, YC3.60 dramatically change the polarization angle of emitted fluorescence upon calcium binding when illuminated with a linearly polarized excitation light. Thus, I thought that I could make a homo-FRET-based calcium indicator whose signal change could be detected by the polarization angle change. Shinya-sensei taught me that rectifier could not be applicable but I might be able to detect the signal change by putting a certain mask in the back aperture of the objective. At that time, I could not understand how to make such mask. But later, you and I started the collaborative work for homo-FRET microscope, and then successfully constructed nice microscope that used a photonic crystal-based compensator.

I only have this story regarding Shinya-sensei but this is very nostalgic memory for me.

Shin'ichi Ishiwata

The first time I met Shinya-san was in the mid-1970s when Shinya-san came to Nagoya University asking my mentor Fumio Oosawa (who passed away at the age of 96 in March this year). Shinya-san found that microtubules in cells depolymerized when cooled, and repolymerized reversibly when warmed, and wanted to discuss this phenomenon with Oosawa-san.

As a graduate student at the time, I was shocked by the brilliant phenomena and the ability to artificially manipulate cell structures. After that, we used Shinya-san's low-temperature treatment method as a method of eliminating the spindles in the HeLa cells in metaphase and regenerating them in any orientation (unpublished results).

The next thing I remember is that Shinya-san “communicated” our paper, which was published in PNAS in 1999. I orally presented the contents at a commemorative symposium held at the Nara Hotel when Shinya-san was awarded the International Prize for Biology. This is also a strong and memorable event.

By the way, Kazuhiko Kinoshita, Jr., my best friend since my undergraduate years, respected Shinya-san very much, and Shinya-san also evaluated Kinoshita's microscope technology. When Kinoshita invited Shinya-san to his home, I was invited to share dinner. On the way back to home, we talked about the spontaneous oscillatory contraction phenomenon of striated muscle that we found. Then, Shinya-san suggested that the phenomenon may be the same as that presented by Dan Kiehart at some academic conference. The abstract of his conference presentation could be cited in a review paper we published later.

Shinya-san was already very famous as an inventor of the Shinya scope when I started my research life in 1969. In addition, the stunning research that captured the dynamics of cytoskeletal structure with Shinya scope has long been a dream of young biologists using optical microscopes.

I pray from the bottom of my heart that Shinya-san's soul may rest in peace.

Letters to Shinya from Colleagues in Japan

Hajime Takahashi

Dear Shinya: It has been 20 years since I developed the Centrifuge Polarizing Microscope with you, but I still remember it clearly. "Capturing the specimen on the disc rotating at 10,000 RPM clearly. " That was your requirement for CPM. I was very skeptical when I started developing this system, but I was very moved when the image was first captured by your outstanding advice and ideas. On the other hand, a powerful laser for illumination and a disk that rotates at an ultra-high speed of 10000PRM are very dangerous, so I was very worried that you would be injured while observing the specimen.. Nonetheless, there are many academic papers, and I am really happy to develop CPM with you.

It was also a wonderful memory to meet Ted and his wife and to have dinner with my wife. Thank you very much from my family.

I am very disappointed to hear this news because I believe you could have continued to be an active researcher until you were 100 years old.

I want to send love and prayers.

Hanako Hayashi

Dear Shinya Inoué,

I am a very lucky person because I had a chance to learn your fantastic science.

Here, I would like to write about my memories with Shinya-sensei.

During my PhD course, I was studying the semi-open mitosis of *C. elegans* embryos and I was interested in the variety of mitosis among eukaryotes. I found your works in which you clearly described the *Barbulanympha* spindle and nuclear envelope dynamics during chromosome segregation with Shinyascope. I never forget how I was excited by your images. After that, I luckily had a chance to attend your lecture in Tokyo in 2010 and we discussed the possibility that protists might show mitosis which we have never seen before. Even now, I am trying to understand the variety and commonality of mitosis in eukaryotes. I am studying the chromosome segregation in a protist, *Trypanosoma*, which has a unique set of kinetochore molecules that are not homologous to the conventional ones. Thanks to Sylvia-san, Tani-san and Noda-san, I had a chance to meet Shinya-sensei again last April at Shinya-sensei's home (photo below) and showed him my microscopic images that captured mitotic spindle microtubules and kinetochores in *Trypanosoma*. Shinya-sensei did not speak much but I remember his smile when he saw my microscopic images. It was really precious time for me.

It was lovely sunny day with the tweet of his birds at his home.

Thank you very much for telling us how wonderful the biology is. May you rest in peace.



