Subtitles and transcriptions

Subtitles and transcriptions are available for selected materials for purpose of helping users understand the contents of the educational sessions.

Uncertain words have been indicated with ?? before and after the part. Parts that could not be understood at all have been indicated as [Audio Not Clear].

Every effort has been made to faithfully reproduce the audio of the sessions as recorded. However, no responsibility is accepted for mistakes or omissions. ESO does not endorse any opinions expressed in the presentations.

e-Session n 617003 - 14th June 2021

The pathology of sarcomas

Prof Dei Tos: So, welcome. First of all, I wish to thank the European School of Oncology for inviting me. I am Paolo Dei Tos, I am a Professor of Pathology at University of Padova. And the topic of my talk will be a summary of the processes behind the diagnosis of soft tissue sarcomas. As you may know, soft tissue sarcomas represent a heterogeneous group of rare malignancies. The overall incidence is of five cases per 1000 people a year. And it is an aggressive disease, as you can see here, 20 to 30% of cases will recur locally, and up to 50% of cases will metastasize. And this happens most often to lungs. The five-year overall survival varies between 55% and 65%. Most of the time, regardless of stage and histology. One of the big issues when dealing with soft tissue sarcomas and also, with the rare cancer in general, is that both rarity and heterogeneity affect diagnostic accuracy. You have to imagine that approximately 30 to 40% of cases which are not diagnosed, I mean, in expert centers in a way get diagnosed erroneously. And pathologists are fully aware of the challenges. That's why sharing cases, asking for a second opinion, working within networks of expertise may all represent a way to overcome the challenge of a diagnostic accuracy in sarcomas. So, the problem is why we want a correct classification, this is quite obvious. It is because pathologic classification represents the rationale of clinical decision making. Of course, it is diagnosis but also prognosis and also biomarker of prediction of response. So, prognostic and predictive biomarkers. You have to remember as a non-pathologist that, in particular, in this era in which molecular genetics is getting more and more important, that, nonetheless, conventional morphology which means the use a microscope, remains a powerful tool. Of course, this will be integrated with immunohistochemistry which is the diagnostic standard. And increasingly, we are implementing molecular genetics, which is very much helpful in selected situations. So, sarcoma classification has evolved through the years, is still based on morphology, and uses a histogenetic approach. We know that these aren't real. Histogenetic means some kind of resemblance to a somewhat normal tissue. So, for example liposarcoma, well-differentiated liposarcoma will be very much similar to normal fat but this, of course, does not represent the process of carcinogenesis. So, all cancer differing shapes works to some extent as it would in normal tissue but they all come from primitive, indistinct mesenchymal stem cells. But this is the genetic approach, is quite practical when you have to set up a classification based on microscopic observation. Of course, classification has evolved with the progressive inclusions of immunohistochemistry and classic and molecular genetics. And all this work is aimed to recognize something that is morphologically as well as clinically distinct entity. And because we should never forget that the main reason to classify neoplasm is just to provide clinicians with the cohesive rationale group of lesions that will

require specific actions. So, I've been part of the WHO panel for the last 20 years. This is one of our first meetings in Lyon, back in 2002 with many of the world leaders in the field of sarcomas. Then we met again in Zurich, in 2013. And the last edition of the WHO was finalized in May 2019, again, back in Lyon. And then, as you may know, in 2020, we finally published the latest edition of the WHO, that you can see here the three fascicles that have been evolving through the last 20 years, two decades of work in the field of sarcoma classification. Just trying to summarize the major changes that occurred through the last two decades. Well, first of all, we have now a definition of tumor category, both in bone and soft tissue which I will teach about shortly. Genetics is now part of some of the tumor entity definition, like for example, MDM2 amplification in well-differentiated liposarcoma and dedifferentiated liposarcoma. Very trained lesions in the '80s and the '90s, like malignant fibrous histiocytoma, MFH, and hemangiopericytoma, that altogether represented up to 70% of sarcoma diagnosis until the late '90s has been totally abolished, which means that you have to find better names, better labels for at least 70% of the diagnosis that we used to do. Some new groups of lesions have been implemented, particularly, in the category on undifferentiated sarcoma and new entities come in. Of course, now, one of the very good things is that for example, gastrointestinal stromal tumors are described in the same way in the different fascicles of the original classification. So, if you open the GI fascicle, will be exactly the same chapter of the soft tissue fascicle. Neural neoplasms are now in the soft tissue and AFX, typical fibers and derma has not been forgotten. That happened some years ago. Then, among some major advances, now we recognize that when dealing with low-grade chondrogenic neoplasm, low-grade chondrosarcoma in the limbs based in the bone of the limbs. As the treatment match that of an enchondroma, you are allowed to use a less aggressive terminology like atypical chondrogenic tumors because at the end of the day there will be curitage. Importantly, some groups of Ewing-like lesion are now a separate entity but I will describe this in more details. Also, these emergent entities, which is all related to the development of anti-NTRK target therapy which represents a group of NTRK related lesions that in a way are interesting in terms of possibility to treat them with specific drugs. Of course, when you get into the description of all the classification, one main question is why don't you move to a molecular classification of cancer, in general, which is probably possible nowadays only for central nervous system neoplasm. But of course, knowing IDH1 status is probably more relevant than the subtle differences between the diagnosis of oligodendroglial neoplasm. But for most of the kinds of cancers, classification is based on morphology. And in this context, genetics play a relevant role but is in a way, factorized within the morphologic features. So, for example, one of the discussions, ongoing discussion is if you have extra-skeletal myxoid chondrosarcoma, which is a mesenchymal malignancy in the limbs of young adults that tend to metastasize usually, 10 years from onset of the lesion, it is necessary just to change the name into a more modern NR4A3-rearranged sarcoma. Well, the answer at the moment is no. No, because all the label, even if we know this is not a chondrosarcoma, it doesn't matter, perfectly describes an entity, of which we know the morphology, the molecular features and the outcome and the natural history. And as you know, in the future, it is quite likely that NR4A3 may be actually found in unrelated lesions. So, we now know that molecular aberration shows some degree from very low to very high gut promiscuity. So, in this context, we think that is better to stay at least for this lesion, for many lesions in that, just stick to the old terminology. Certainly, the introduction of next generation sequencing or better to say, massive parallel sequencing, which represents high-throughput sequencing technologies which is just fast, robust, highly sensitive with a broader range of applications from genomics, transcriptomics to epigenomics. Of course, has changed significantly in the field. But of course, what is important to understand is that its application needs to be played in, again, in concert with morphology. Now we'll give you later on some examples, but certainly, MPS has contributed significantly to validate classification, to identifying newer diagnostic markers. For example, that happens with STAT6 in solitary fibrous tumor, STAT6 is up-regulated because of fusion gene that included STAT6 with NAB2. So, currently, is easier, instead of looking at the fusion gene, to look at the overexpression of the STAT6 protein which represents the perfect confirmation for morphological diagnosis of solitary fibrous tumors. New tumor entities, like the CIC and BCOR sarcomas, I will touch base shortly. Elucidation with sarcoma pathobiology, now we knew why the amount of fibrosarcoma protuberance can progress to a fibrosarcoma variant, the

one that can rarely metastasize to the lungs. And also, of course, in the story of NTRK in GIST and beyond is so emblematic, the identification of new potential therapeutic targets. So, for example, pseudomyogenic hemangioendothelioma is a mesenchymal neoplasm occurring in most of the time in the limbs of adult patients with a very distinctive involvement of different tissue planes. Despite that, the course is very indolent. And those patients presenting in one limb with multiple lesions, occurring in the skin, in the deepest of tissue, even the bone, actually, we know now they don't deserve amputation, as most often happened in the past. We know that multiple nodulectomy will in a way guarantee a control of the disease, of course, preserving the function of this patient. That's why it's relevant to recognize this entity. And having the information that all these cases that have been recognized morphologically actually do have a specific fusion gene represented by the combination of SERPINE1 with FOSB. Of course, assume many relevant confirmed findings that we are really dealing with something unique. And in addition to that, just to give you an example as MPS, in a way, can have picking up new diagnostic marker. Well, the fact that FOSB is up-regulated will allow us to demonstrate its presence in the nuclear neoplastic cells with immunohistochemistry. So, nowadays, to make a diagnosis in psuedomyogenic haemangioendothelioma, in addition to the clinic, to the morphology, to the immunophenotype, we don't need actually to go through genetic analysis. We simply mean one immune...in addition to cytokeratin in a vascular marker which is represented by FOSB. New tumor entities also represent an important example of correct application of molecular genetics. And actually, 15 years ago, more than 15 years ago actually, we were quite puzzled by tumors that seem to belong to the family of Ewing sarcomas which, as you know, the prototype around sarcoma is high-grade occurring when dealing with pediatric groups of patients, mostly in the bone. But we know that they can occur in the soft tissue, in the skin, at the visceral sites. And when this happens, there is no predilection for young person. They can occur any time during the life of the patient. Well, few years ago, exactly among this group of un-perfect tumors, un-perfect because CD99 was not so much well-expressed. And CD99 is usually strong and intensely expressed, strongly and intensely expressed by Ewing sarcoma. And the genetic was not perfect because basically EWS gene was not rearranged and the clinical was not because those patients actually tend not to respond so well to the Ewing sarcoma systemic treatment. Well, actually we know now why? Because they were not Ewing's at all. But they were, for example, cases of CIC-DUX4 fusion positive round cell sarcoma. And this is the old tool, which is represented by cytogenetics that can demonstrate the chromosome involved with exchange of genetic material. And then, of course, molecular genetics can mean particular NGS that help a lot in identifying this new category of lesions. Which now, despite the fact that extremely rare, well now we managed to recognize them. There is a slight male predominance as you can see the pick-incidence in the third decade. They are found common soft tissue than bone. And their form varies, it is a little bit misleading, so, it's not the perfect round cell sarcoma but it can show spindling, it can have epithelioid morphology, it can have myxoid change. All things in a way, represent a major diagnostic challenge. And of course, these cases do have a specific genetic fusion that we can identify in order to support our diagnosis. These are some of the cases that looks a little bit Ewing's for the non-pathologist. When you look at this horrible size here, much bigger, very hyperchromatic. These is not what you observe usually in Ewing sarcoma. That's why 15 years ago, we tried to single out those cases of so-called atypical Ewing's that actually were not Ewing's. Some other examples of CIC rearrange sarcomas are not round, are epithelioid or spindling. Sometimes, actually most of the times, in keeping with extreme aggressiveness of the lesion you have abundant necrosis, which means that the balance between tumor, growth rate and vascularization is that unbalanced that part of the tumor die. But we know that this actually will present a negative prognostic factor. We may use immunohistochemistry, of course, in order to make the diagnosis to support diagnosis. Because CD99, as I mentioned to you is variable, patchy, not like Ewing's. We get WT1, ETV4 and NKX2.2 which is a nice marker, a recent marker for Ewing, it tends to be negative. So immune and morphology, we can help. I'm saying that underlying this because if you go to the WHO classification, WHO doesn't regard molecular genetics as mandatory, even when dealing with CIC rearranged sarcoma. But the reason is that WHO stands for anybody in the world. And of course, access to fusion, gene molecular analysis is very limited in up to 70% of our planet. Then of course, things evolve. So called CIC sarcoma actually are growing. So,

these CIC-NUTM1 family of sarcomas, but I'm using this just to try to share with you some of the challenges when you start describing new things. Because as you know, NUT neoplasm has been invented in Boston and actually, by Chris French, at Brigham and Women's, with the NUT. Another one, actually, you know is it can be a target. Actually, what's in France may be represented like a NUT CIC-rearranged sarcomas, actually, from the other side of the ocean, actually represents one of the cancers that shows NUT-rearrangement. So, you know, when you start classifying things based on molecular genetics, as you can see, sometimes there is a limited improvement because, for example, some of the debates around this specific entity is very far from being settled. The second group of these new category within WHO, of format atypical Ewing represented by BCOR which is round cell sarcoma, which shows a male predominance with a peak in the second decade of young patients. Far more common in bone and soft tissue. And here, is funny because in Ewing, we tend to classify this lesion among round cell sarcoma but most of them actually are spindle cell sarcoma. As you can see here, some myxoid fitting, some spindling of neoplastic cells. And this is interesting because also BCOR does not respond so well to systemic treatment. And this is one of those rare examples that we have of post-chemo, because sarcoma and you can see here, most of the tumor cells get around in cluster or in fascicles, actually are absolutely vital. The tumor didn't respond at all to the cytotoxic treatment. And BCOR, of course, can be recognized based on immunohistochemistry when you have most of the time fusions in between BCOR, Cyclin B3. You can either choose BCOR or Cyclin B3 in order to pick up the up-regulated protein and support your morphologic diagnosis. But the question is, why do you create these new entities? Why do you really need sub-segmentation, such a granular approach to that so-called round cell sarcoma? Well, the reason is clinical, as you can see, BCOR behaves far differently from Ewing's and from CIC sarcoma, CIC sarcoma being the most aggressive. This, of course, doesn't mean that you have to treat them differently at the moment because the number are so low that we don't have clinical studies demonstrating different outcome using different treatments. But this is the only way in order to provide clinicians with the evidence that maybe in the future they will be managed to create clinical trials, try to, in a way, identify better treatments based on the specific histologies. And within this group of new entities also belong the so-called NON-ETS fused sarcomas. Which means that situation in which the EWSR1 gene, or the first gene that molecularly is very similar, will fuse with NON-ETS related genes but with different genes, in particular NFATC2, PATZ1. And that, of course, is an EWSR1-NFATC2. And molecularly, this is very fascinating. We were the first to report this problem a few years ago, where we saw that EWSR1 in some tumors and not only rearranged but also amplified. And this is a very typical lesion, which is, as you can see here, is very rare. It occurs exquisitely in the bone. You can see the x-ray of this lytic lesion. And this is the morphology, which is again challenging because this could easily be a metastasis of a carcinoma because the neoplasm looks like a cancer. But the patient is very young. This is a one single lesion. The immunophenotype in molecular is that of EWSR1-NFATC2, primary sarcoma of them. And this is some of the recent papers, the new way draws attentions on this new entity. As it is, now, the example that we just mentioned. This is a male of 19 with the mass in the thoracic wall with lung metastases onset. And the thoracic wall is the perfect place for these new group of EWSR1/PATZ1 tumor that, by the way, shows a poly-phenotypic phenotype. Which means they show differentiation with this means that is a myogenic marker, S100, which is a monocytic or for example, neuro marker. So, this is a group of new fascinating neoplasm. Of course, we start now to recognize, hoping them to get more and more information of nature of history of these diseases. And then going to the conclusion, one important piece of information comes through identification when new potential targets, few years ago, just because we were interested in so-called wild-type, GIST. GIST that do not have mutation of KIT, PDGF alpha or neurofibromatosis one gene or RAS, BRAF genes. We touched base on the case of typical GIST but showing a ETV6 factory, which is exactly the same gene fusion that you have, for example, in infantile fibrosarcomas. I know that now some people are trying to say, okay, some, now, of them NTRK rearranged lesion within the GI tract and not actually GIST, but this was a GIST, with KIT and DOG-1 expression and [Audio Not Clear]. So, I'm not denying that this new NTRK related entity may occur in the GI tract but wouldn't simplify saying that these are not GIST because they have NTRK and no other, and not mutations with classic KIT and PDGFR genes. Anyway, at the time we didn't know anything about NTRK inhibition. Of

course, now is a very, very important thing. You can see the data from one of the two drugs which are currently available with the fantastic waterfall plot analysis in which you see how much response you see particularly in patients having NTRK rearranged neoplasm, in particularly sarcomas, like infantile fibrosarcoma or others recently introduced entities like lipofibromatosis-like neural tumor and this also new category of tumors that closely resembled peripheral nerve sheath tumors. This is one example. Why we call L-LNT? Because it looks like lipofibromatosis. But in addition to that, sheer expression S100, CD34, and NTRK and they have most of the time, in fact, one gene rearrangement. And this is also true for the tumor resembling peripheral nerve sheath tumors. Whereas L-LNT is most of the time it's benign even if it can grow really big. So, you may want to downsize the lesion with the use of these new drugs. TRPNST actually varies from benign to very aggressive, depending on the morphology. So here, morphology is the biomarker predictive of clinical behavior. And it's just to share with you the immune. This is S100, and this is, of course, NTRK that in a way when expressed strongly, always need in the case of sarcoma to be checked where the molecular genetics. Of course, those lesions are extremely rare. You also have to remember that infantile fibrosarcoma responds pretty well to conventional drugs and/or radiotherapy, with an overall survival at five years of 95%. And the others subtypes, some are benign, some are not clearly defined. So, this is why WHO decided to present these new cases, like an emerging entity which needs farther validation in the future. That's why within the sarcoma community which is a group of friends who are very passionate about sarcoma, very different expertise, like surgery, chemotherapy, I mean, medical oncology, orthopedic oncology, pathology, molecular biology. We decided to look into that and say, okay, how do we pick up these patients? Knowing that there exist in a way several, several possible analytical meters that we can use. Also, of course, we may go, okay, we do NGS in all the cases, we got to interview and people try some time to do this. And of course, the yield is ridiculous low because the lesions are so rare. So, one good compromise, maybe, is using immunohistochemistry when the morphology and the immunophenotype, which means S100 and CD34, in a way raise the suspect and if NTRK immuno, upon NTRK immuno is positive then try to confirm with molecular genetics. This is a reasonable approach in order to avoid to leave patients behind which of course is ethically unacceptable. Then, of course, we have to know that the more genes you test, the more things you find. So, for example, you get into this MGA-NUTM1 new subtype of high-grade spindle cell sarcoma. Which means one case, two cases, three cases, this is kind of a thing that I don't personally like so much, because this, of course, tend to create some kind of impossibility to really know what these lesions represent. So we don't want this type of reaction among patients and among clinicians, but particularly because you have to know this has been demonstrated recently by the very nice, very good molecular geneticist in London, the one who invented the genetic of sarcoma. The more NGS you do the more gene fusion you find just as a stochastic event. And so, in the vast majority of those fusion genes are not a driver of sarcoma genetics, simply passengers. That's why you don't need to create one entity all the time you pick up a new fusion in a given neoplasm. And with Paolo Casali and Alessandro Gronchi, which I also do reasonably to be on... What are the conditions in which you create a new entity? Each is a combination of different things. It is not just a new cluster which is defined morphologically or by immuno, by molecular genetics. Even the simple presence of predictable biomarker should not be regarded enough. You need a clinical context, you need a morphology and molecular genetic correlate, the therapeutic core. Then you have an entity, but you have just to single out one of those factors. You just have a description of things that in a way the recognition, which is not adapt for it yet. And among medical oncologists there is a tendency to believe, okay, we don't really need pathology anymore because we can't sequence everything. And then, you pick up all the entities you want. You, in a way, improved diagnosis. Well, certainly as happening in these abstracts, if your sarcoma NOS diagnosis comes in up to one fourth of your diagnosis, I don't think you need it. And just plot on. You need a good pathologist. In particular, when you give an example of a changing diagnosis of a leiomyosarcoma to liposarcoma based on genetics, this is something that a first-year resident here in Padova will never do. They would recognize it easily. So, NGS is not the answer to medical ignorance. And in fact, there is no real evidence actually that sequencing all the cancers has any clinical benefit at the moment. And I'm not against the fact that when you fail two different lines of therapy, you can interrogate

yourself and the tumor, say, okay, is there anything in the tumor that we can find and we can match with some therapy? This is a very reasonable, this is something we do. And I think we should do but this is another story, that's saying, okay, we don't need to contextualize a lesion anymore. We can go pathology agnostic, and simply look at the genes because it creates problems like in this patient with a Carcinoid the diagnosis of which was translating into Ewing because of the presence of EWSR1 ERG fusion. Well, then, a bit later on, we reported in Nature the fact that actually costing now that can have those types of fusions because as I mentioned to you before gene fusion, genetic abnormalities show a high degree of molecular promiscuity. And actually, if you want to follow this idea look at the most recent papers in which they try to have NGS within this platform in order to say, okay, what does change? Well, if the information is that, now we know the importance of the TP53 signaling pathway in sarcomas. Actually, we published that, 25 years, ago using much less powerful technology. So, it is now a great step forward. There is another paper from more-or-less the same group of people in which they conclusion are our data suggest that molecular profiling should not be using routine practice. But of course, you always say that, once further exploration in clinical trials, which means that we shouldn't sit around a cave, forget about pathology, go pathology agnostic, do NGS will be happy. This is not their reality. So, my conclusion is, certainly soft tissue tumors represent a global challenge. We know the rarity affect diagnostic accuracy; molecular testing, despite some of the criticism I raise is actually very useful in cancer morphology. I, however, believe that excessive molecular segmentation is potentially confusing as it stands now. And classification is important. I'm sure that WHO classification and its distribution, its diffusion among the community of pathologists is crucial to improve quality of pathological diagnosis. And this is the end of my talk and I wish to thank you for your attention.