The Cappadocia mesothelioma epidemic: its influence in Turkey and abroad

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Abstract: The epidemic of mesothelioma in Cappadocia, Turkey, is unprecedented in medical history. In three Cappadocian villages, Karain, Tuzkoy and "old" Sarihidir, about 50% of all deaths (including neonatal deaths and traffic fatalities) have been caused by mesothelioma. No other epidemic in medical history has caused such a high incidence of death. This is even more unusual when considering that (I) epidemics are caused by infectious agents, not cancer, and (II) mesothelioma is a rare cancer. World-wide mesothelioma incidence varies between $1/10^6$ in areas with no asbestos industry to about $10–30/10^6$ in areas with asbestos industry. This article reviews how the mesothelioma epidemic was discovered in Cappadocia by Dr. Baris (my mentor), how we initially linked the epidemic to erionite exposure, and later (with Dr. Carbone) to the interaction between genetic predisposition and environmental exposure. Our team's work had an important positive impact on the lives of those living in Cappadocia and also in many genetically predisposed families living around the world. I will discuss how the work that started in three remote Cappadocian villages led to the award of a NCI P01 grant to support our studies. Our studies proved that genetics modulates mineral fiber carcinogenesis and led to the discovery that carriers of germline *BAP1* mutations have a very high risk of developing mesothelioma and other malignancies. A new, very active field of research developed following our discoveries to elucidate the mechanism by which *BAP1* modulates mineral fiber carcinogenesis as well as to identify additional genes that when mutated increase the risk of mesothelioma and other environmentally related cancers. I am the only surviving member of this research team who saw all the phases of this research and I believe it is important to provide an accurate report, which hopefully will inspire others.

Keywords: Erionite, *BAP1*; environmental mesothelioma; gene and environment interaction

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Mesothelioma caused by mineral fibers

Mesothelioma is characteristically associated with asbestos exposure. Asbestos is the name that was used in the 1970s to refer to six commercially used carcinogenic mineral fibers: chrysotile, crocidolite, amosite, actinolite, anthophyllite, and tremolite. Millions of people were exposed. The use of these fibers were regulated in most of the world during the past century, and it is either entirely prohibited (Western Europe) or tightly regulated (i.e., some countries, like the USA, still allow the use of products that contain chrysotile, which is considered a less dangerous form of asbestos). In Turkey, regulations imposed in 2005 restricted the use of asbestos, but the production and processing of asbestos products was entirely banned only in 2013. In addition to the six regulated asbestos minerals, there are about 400 additional non-regulated fibers that possess physical and chemical structures similar to commercial asbestos and which may also be carcinogenic (1,2). In recent years, some of these fibers, such as erionite and antigorite have been shown to be at least as potent as asbestos fibers in causing human mesothelioma, while others, such as palygorskite, were found not to be oncogenic (3-5). Some of the non-regulated fibrous minerals have been used commercially in

recent years. In addition, in areas where deposits of fibrous minerals are present, erosive processes often enhanced by human activity disturb soil, releasing mineral fibers into the air (2,6,7). Once fibers are dispersed in the environment and become inhalable, individuals may be exposed, and over time may develop mesothelioma (8). In the US, and particularly in the more arid portions of Nevada and California, both natural dust emissions and human activities such as off-road-vehicle (ORV) use, road construction, and development of rural areas have significantly increased emissions of mineral fiber-containing dust (9-11), increasing the risk of mineral fiber-related diseases. However, in none of these instances, an epidemic of mesothelioma of the proportion even close to what has occurred in Cappadocia has been documented.

The association of mesothelioma with asbestos exposure is well established (12). The mean age at diagnosis is 74 years old, and due to occupational exposure, the male to female sex ratio is 4:1–8:1 (highest in areas where asbestos exposure is high). The latency period of asbestos-associated mesothelioma is on average 30–60 years (13), which accounts, at least in part, for the fact that malignant pleural mesothelioma (MPM) incidence is still rising worldwide despite working bans on the use of asbestos in the early 1990s (14). Only a fraction (about 5%) of those professionally exposed to asbestos for many years develop mesothelioma (3,8,15).

Asbestos-related mesothelioma in Turkey

Turkey does not have a National Mesothelioma Registry. The Cancer Control Department of the Ministry of Health published cancer statistics in 2015. According to the statistics, age standardized incidence rate of mesothelioma was reported as 1.1/100,000 for males and 0.6/100,000 for females (16).

The first epidemiological surveys of mesothelioma in Turkey and its relationship to asbestos were performed by my mentor, Professor YI Baris and published in Turkish (17). Baris had become fascinated with the disease when he worked as a medical officer at the Naval Medical Hospital treating sailors who had developed mesothelioma. In 1969, Baris resigned from the military and became the Chair of the Department of Chest Diseases at the University of Hacettepe at Ankara until 1998. I started working with him during my residency program and entered his department in 1989 and subsequently held that position from 2012 until 2015.

Baris decided to study mesothelioma because he realized there was limited information about this disease and the extent of asbestos exposure in Turkey. He established a collaboration with a geologist Dr. Asim Goktepeli, investigating central Anatolia, where there were many asbestos mines. Their first field trip was to the town of Mihaliccik in October of 1973, with a population of about 5,000 people. The local physician told Baris that there was no mesothelioma in the region, only many patients with tuberculosis. Upon reviewing the X-ray films of the "tuberculosis" patients, Baris realized that many of these patients had pleural plaques, not tuberculosis, and were receiving ineffective therapy. During the same field-trip, Baris and his collaborators visited a nearby asbestos mine where workers were using asbestos without any precautions. The people in that town and in the nearby town of Eskisehir were also using asbestos products without precautions. That field trip convinced Baris that there was a major problem with asbestos in Turkey, and he took on the mission of studying asbestos exposure in Turkey to help prevent mesothelioma. Baris went back to Mihaliccik each of the following weekends. After about 2 months, he had reviewed 3,880 chest X-rays from Mihaliccik taken in the previous years. He found that 10% of the patients had pleural plaques, and that six patients had mesothelioma. From that time, Baris and his team spent almost every weekend on road trips to all of the villages in central Anatolia, surveying for asbestos exposure and mesothelioma.

Baris did not want to ask permission to the University for these trips, as he disliked the administrative requirements associated with an "official" field trip. He was an independent spirit and very much a military man, used to giving orders and being obeyed. He did not want any administrator to have control over him. Thus, he traveled on his own time, with his own money, and never asked for reimbursement. We, his associates, were required to travel with him under the same set of circumstances. We did it because his passion for research to save human lives from asbestos exposure and mesothelioma was contagious, but also, in all honesty, we had no choice. Professor Baris was the boss and he expected nothing less from us than obedience. Because we had no research funding for this work, we usually slept in the car; although over time the villagers got to know us and hosted us. During these field trips, we worked continuously, seeing one patient after the next, taking X-rays and developing them on site. We arranged for a portable X-ray machine to travel with us and we paid the technician out of our personal funds. Finally,

late in the evening when the work was completed, we would rest and have something to eat. Our work was similar to a military campaign.

From 1969 until the mid-1980s, under the guidance of Baris, we surveyed almost every village in the vast region of Anatolia (18). Our studies demonstrated the presence of asbestos, pleural plaques and mesothelioma, as well as other diseases and malignancies in many patients (19-21). Baris's studies provided the first reliable medical and scientific information about the extent of asbestos exposure and related diseases in Anatolia. This gave the Turkish health authorities the information upon which to act to implement preventive measures to reduce asbestos exposure and educate the population about the risk of asbestos exposure.

Erionite-related mesothelioma in Turkey

As we were conducting these studies, a colleague, Dr. Ozerkan, told us that he had been to a Cappadocian village called Karain where the villagers claimed there was a high incidence of mesothelioma. Dr. Baris wrote a letter to the headman of Karain and invited him to come to Ankara and bring all information (X-rays, etc.) for review. The headman showed up in our department accompanied by a patient who was being treated for tuberculosis. Baris admitted the patient to the hospital and we diagnosed malignant mesothelioma. We obtained and reviewed the files of all patients that in previous years had been admitted at Kayseri Hospital, the closest hospital to Karain, for chest diseases. Almost all of them had been treated for tuberculosis, but according to the Karain headman all of them had died within a year, in spite of treatment.

This review was enough to convince Baris to pay a visit to Karain. In October of 1975, the team of physicians, with a technician carrying a portable "Picher" X-ray machine, our colleague geologist Dr. Goktepeli, and under the guidance of our Chief, Professor Baris, drove all the way to Karain, a 5-hour drive from Ankara. Working around the clock, we took X-rays of about 200 people over the age of 20. The population of Karain at that time was about 800. The X-rays films were dried by hanging them over ropes that were normally used to hang clothes in the back yard of the Karain Health Center. We diagnosed 12 patients with probable mesothelioma, all diagnoses later confirmed by chest biopsy and histology. The incidence of mesothelioma in Karain was about 1,000 times higher than in any other village we had surveyed in central Anatolia. Equally surprising, our geologist assured us that there was no excess of asbestos in Karain compared to any other village we had surveyed, a fact later confirmed by multiple international studies (7,22). Moreover, in Karain the incidence of mesothelioma was about 1:1 between men and women, and 1:1 between pleura and peritoneum, something highly unusual because mesothelioma is much more frequent in the pleura of men, at least among people exposed to asbestos (5). Moreover, the mesothelioma patients were in their early 50s, almost 20 years younger than the mesotheliomas we had found in other villages. As we were contemplating this very unexpected set of findings, we learned of another town located 40 km from Karain, where according to the local physicians there was a high incidence of "tuberculosis"—Tuzkoy, a larger village that at the time had about 3,000 people. When we visited Tuzkoy the following weekend, we were confronted with a mesothelioma epidemic of similar proportions to Karain. Later, we made a similar finding in the village of Sarihidir located between Karain and Tuzkoy. Intriguingly, in nearby villages, for example the village of Karlik situated 3 km from Karain, we did not find any excess of mesothelioma. These findings, first reported by Baris in *Thorax* in 1978 (23), started a flurry of investigations by several international research teams, into the causes of this mesothelioma epidemic. Erionite, a fibrous mineral, was found in the lung tissue of affected patients, and in the air of these three villages. Animal experiments demonstrated that erionite was more potent than asbestos in causing mesothelioma, and it was concluded that the epidemic of mesothelioma in these villages was caused by the presence of these fibers in the stones used to build the houses of these villages and in the gravel used to pave their roads (7,21,22,24-27).

Genetics and mesothelioma

It is now well accepted that genetics plays an important role in mesothelioma, especially in certain families. This is an active and growing field of investigation. The hypothesis that genetics caused mesothelioma was conceived and later proven by studying the mesothelioma epidemic in Cappadocia.

During the years since Baris's initial discovery of a mesothelioma epidemic in Cappadocia, we accompanied several distinguished visiting scientists to these villages for brief visits, as most were afraid of erionite exposure, and we conducted research with some of them. Although we had linked the epidemic to erionite, we were puzzled by the extent of the mesothelioma epidemic: over 50% of

all deaths in these villages were caused by mesothelioma, an incidence that exceeded any reported incidence of mesothelioma in workers professionally exposed to asbestos. Could there be something more in addition to erionite responsible for this epidemic? I brought to Baris's attention a paper by Dr. Michele Carbone reporting the presence of simian virus 40 (SV40) DNA in some US mesotheliomas. This virus contaminated polio-vaccines, thus millions of people had received infectious SV40 over the years as they got vaccinated with polio (28-30). I convinced Baris to invite Carbone for the Turkish Thoracic Society meeting in Ankara in November 21, 1997. Upon Carbone's request, we drove for a 2-day trip to the Cappadocian villages before the meeting with Dr. Mutti (a renowned mesothelioma expert), Dr. Umran Dogan and Dr. Meral Dogan, two geologists who had replaced the retired Dr. Goktepeli.

Carbone, at 37 years old, was much younger than we expected. He was very direct and forthcoming and asked many probing questions to Baris. Then he would ask us if we agreed with Baris. Carbone did not seem to understand that whatever Baris said we had to agree with him and that was the way we worked. Disagreements were not allowed, especially in front of a visitor. When he noted and asked Baris why some families in Karain and Tuzkoy appeared to have many more cases of mesothelioma than others, Baris answered that their homes had more erionite than others, or maybe a more dangerous type of erionite. We were in Tuzkoy. Carbone said he did not believe it because everybody in the village was exposed to the same dust. He turned to Dr. Umran Dogan and asked him to take samples from a house where everybody had died of mesothelioma, one of the so-called "houses of death", where we had found several playing children covered with erionite-dust (*Figure 1*), and from a nearby house where apparently nobody had died of mesothelioma. Baris decided he had had enough and basically told Carbone he did not know what he was talking about. Dogan however, went ahead and took the samples.

That was a defining moment of the research. Baris and all of us knew that Carbone's objection was valid. We had already noted and discussed among ourselves the unusual prevalence of mesothelioma in certain families and the fact that women who married villagers in Karain and Tuzkoy and then moved into these villages rarely got mesothelioma, but we had no explanation for that. We were conducting a study to investigate the potential links between the occurrence of mesothelioma and the presence and distribution of human leukocyte antigens (HLA) in

Figure 1 Tuzkoy Nov 1997: "house of death" (left), Dr. Dogan, (right) Dr. Carbone and children we found playing inside the abandoned home.

patients environmentally exposed to asbestos and erionite in rural Anatolia. That study, published a year later, revealed the HLA-B41 antigen in 19.4% of the patients compared to 0.8% of the healthy controls among Tuzkoy inhabitants (31). However, Baris was not about to discuss uncertainties and hypotheses with a visitor, and got annoyed when Carbone pointed out something he knew he could not explain. At that time, Carbone's hypothesis was that maybe the villagers had received polio vaccines contaminated with SV40. Reluctantly, Baris agreed to conduct the SV40 study, to see if this virus, later shown to be a co-carcinogen with asbestos (32), had anything to do with mesothelioma in Cappadocia. The results, turned out negative (33), and Baris told us he was no longer interested in working with Carbone.

Meanwhile, I was helping a bright Ph.D. candidate from Harvard that I had introduced to Carbone. We were collecting and sending to Carbone all sorts of information about the villages and the villagers. I was helping Carbone, because I knew (as Baris knew) that there had to be something more than just erionite to cause such a devastating mesothelioma epidemic. My intuition was that Carbone had the enthusiasm and the drive to help us discover new information to help the villagers. We realized that there were numerous cases of intermarriage in these

villages, an effect caused by the fear of nearby villagers to marry anyone from the three mesothelioma villages. Nearby villagers believed that mesothelioma spread across families via marriage. These beliefs were dismissed by "us" physicians and scientists, but it turned out that they were correct. Intermarriage had confused the results of previous studies we had conducted, in which we had ruled out genetics, as individuals marrying into several mesothelioma families had developed mesothelioma, a finding that apparently ruled out genetics as a predisposing factor. Now instead we realized that those people were also coming from other mesothelioma families! It made me very proud that our work was accepted in the *Lancet* (34). Later Baris celebrated me for this work. Later I learned that although Baris was not a co-author, Carbone, as a courtesy sent Baris the paper ahead of press. Baris answered he did not believe it, and asked to review all the original data. To Baris's surprise, Carbone immediately sent him all the original data via express mail with a nice note. Baris went to the villages and spent 2 weeks there reviewing all the pedigrees that we had built, which showed that susceptibility to mesothelioma was transmitted as an autosomal dominant character. Now it was my turn to be surprised—Baris sent a note to Carbone stating that he had reviewed all the data and except for two minor mistakes the pedigrees were correct, so, he wrote to Carbone, all of us would like to work with you to discover the "mesothelioma" gene.

Since then, we had several years of wonderful and true collaborative research. We were motivated by one common goal: to help the villagers and defeat the mesothelioma epidemic in our life-time. Baris was getting old, as he reminded us several times—he estimated that he was born "probably" in 1929. We were determined to fix the problem during his lifetime. Baris no longer considered Carbone to be an outsider. He liked and trusted him and referred to him as "an honest man," which was the highest compliment in Baris's vocabulary. We needed funding to identify the mesothelioma gene—assuming such a gene existed, something that most experts at the time did not believe at all. Initially Carbone worked with us during his vacation time as he had no funding for this research, and his Chair did not want him to work on a project in remote villages in Cappadocia. Then, in 2003, Carbone won a grant from the Cancer Research Foundation of America. A year later, in 2004, he won a grant from the American Cancer Society to study the contribution of genetics to mesothelioma in Cappadocia. That was the first time a research grant was awarded to study the novel hypothesis that genetics modulated mineral fiber carcinogenesis. Funding accelerated research, and with the data developed with these grants, in 2006 we published a new paper in *Cancer Research* (22) and Carbone wrote a Program Project grant (P01) to the US National Cancer Institute to study mesothelioma. Carbone's own "Project 1" proposed the existence of a mesothelioma susceptibility gene that he proposed to find during the 5 years of the grant, and included several colleagues in his project: Drs. Cox, Pass, Steele and Yang in the US, and Drs. Baris, Dogan and myself in Turkey. Drs. Mossman and Testa were collaborators with unrelated projects to study other aspects of mesothelioma in mice. Would the NCI fund a $10 million research project to identify a gene in a remote part of Turkey? A gene that at the time most experts did not believe existed at all? One of the reviewers noted that although she thought it was "almost impossible" that we would find the gene because we did not have enough "power", i.e., too few people to study, the reviewer thought the project was so amazing that it deserved a chance. Against all odds, the P01 was awarded to Carbone in the summer of 2006.

During those years, Baris and I travelled together to the villages many times. Often we would set up a table in the center of the village or in the "bar" and Baris would ask everybody to donate their blood (*Figure 2*). When we diagnosed mesothelioma, the patients were surgically treated by our collaborator Dr. Hadi Akay who was the Chief of Thoracic Surgery at the University of Ankara. Even one of his residents had spent time at Dr. Pass's clinic for short-term training. Carbone came several times to the villages. Sometimes he came alone, other times he came with his collaborators: Drs. Pass, Yang, and Steele. Some collaborators declined to come to the villages because they were afraid to get exposed to erionite. During our trips to the villages, many villagers asked for medical consultation for different complaints. We saw them all, never charged anybody and often gave them free medicine that we bought for the purpose of distributing them to those in need (*Figure 3*). Together with his student, Monica Rdzanek, Carbone worked in the laboratory with me at the University of Hacettepe, where he extracted and purified all the DNAs we collected in the villages (*Figure 4*). He was a "Professor"; it was unusual for us and for our students to see a "Professor" working with his own hands in the laboratory.

The gene eluded us for 4 years. We were in the pre-next generation sequencing (NGS) years, so the gene had to be identified by genetic linkage analyses (performed in

Figure 2 Sarihidir, Health Center. Blood collection for ELISA studies. Standing, right, Dr. Emri.

Figure 3 Our "office" in the village of Sarihidir. From left, Drs. Carbone, Baris, a mesothelioma patients and her niece, and Dr. Emri.

Dr. Cox's laboratory) for which we were underpowered, and using array-comparative genomic hybridization (aCGH, performed in Dr. Testa's laboratory), which are not sufficiently sensitive to identify minute genetic deletions or nucleotide level mutations (35). However, these analyses helped us identify areas in the genome that "might" have carried the potential gene mutation. Once these areas were identified, they were manually sequenced (Sanger sequencing) in Dr. Carbone's laboratory. In addition to the Turkish families, Carbone was also investigating US families with a similar high incidence of mesothelioma. These families had learned of our studies in Turkey and contacted

Figure 4 Extracting DNA for SV40 virus PCR study at University of Hacettepe, 1997. From left, Drs. Emri, Carbone, Mutti, and Kocagoz.

Carbone to be included in his studies. We went from great excitement, for example, when genetic linkage analyses identified a very promising area on chromosome 6, to deep depression, when, for example, Dr. Nasu in Carbone's lab completed the sequence of the entire chromosome 6 and concluded there were no consistent genetic alterations. Over the years, miles and miles of DNA were sequenced and nothing was turning up: we knew we did not have enough time left to sequence all the potential hot spots identified by genetic linkage studies. Nowadays, with the development of NGS, the whole thing could have been done in matter of months!

We also had some achievements. In the fall of 2001, during one of Carbone's trip to Cappadocia, we were in a small restaurant along the Red River near Sarihidir, discussing the sad experience of having seen three more new mesothelioma patients. The discussion turned to the need to move the villages to a safer place with no erionite. Many villagers were genetically susceptible to erionite, thus exposure had to be avoided to prevent or at least delay the onset of mesothelioma. Baris had tried several times to convince the Turkish authorities to build new villages and relocate the villagers. His proposal had been received with initial enthusiasm but nothing had come out of it. So Baris was skeptical, he no longer had any contact with the Ministry of Health and did not want to waste more time. By chance, a waiter who had been overhearing our conversation interrupted us and said he had a cousin who worked for the Ministry of Health and offered to arrange for us to meet the Ministry. Carbone wanted to try, and Baris reluctantly

agreed that "if" the cousin got us an appointment he would come with us. We got the appointment. Carbone delayed his scheduled travel back to the US and a week later Drs. Baris, Carbone, U. Dogan and I met with the Turkish Ministry of Health in his office. Baris asked Carbone to speak, as he hoped that a foreign "Professor" might better impress the urgency of the problem. The Ministry listened very attentively to Carbone's presentation that Baris translated for him in Turkish. As we left the Ministry said he would fix the problem.

Soon after our meeting, the Ministry of Health dispatched orders to build a new town for the village of Tuzkoy. A total of 224 new homes were completed by 2004. The villagers moved in the new houses, built with bricks and cement. The Cancer Control Department at the Turkish Ministry of Health played a key role in insuring that the village was built promptly. Later on, the health center, which Carbone also desired to complete, had been finished in the village of Tuzkoy to facilitate research and treatment of mesothelioma patients. Overall, it was a tremendous improvement in the living conditions as the new homes had running water, kitchen, bathrooms—all things that were missing in most of the homes in the old village. There was also a small clinic and a physician assigned to it to help the villagers with their medical needs (*Figure 5*). Following up on the same project, the Karain villagers were moved.

Some critics raised the concern that the "new Tuzkoy" had been built too close to the old village, and therefore they suggested that the villagers continued to be exposed to erionite. This speculation led to numerous media inquiries and caused widespread concern. In parallel studies, Carbone discovered (7) that in North Dakota, over 300 miles of roads had been paved with erionite and he was working with the US Environmental Protection Agency (EPA) to assess the danger of that exposure to the population. They had found erionite fibers in the air of school buses transiting on the erionite roads but there was no published reference to know if those concentrations were dangerous. This offered a terrific opportunity. Measuring the concentrations of erionite in the air in the old village of Tuzkoy—where 50% of villagers developed mesothelioma—and in nearby villages with no mesothelioma and in the new village of Tuzkoy, would address the question of whether there was any risk of erionite exposure in the new village of Tuzkoy. At the same time these data would provide the required reference measures to the EPA to assess if the erionite concentrations in the air of the North Dakota erionite-gravel roads posed a danger to human health. Carbone and

Figure 5 Mission accomplished, 2007. Dr. Baris (left), and Dr. Carbone (right) in the background the new village of Tuzkoy.

a team of EPA scientists led by Dr. Aubrey Miller spent 3 weeks in Cappadocia conducting air sampling. The results revealed that (I) the air of the new village of Tuzkoy did not contain erionite, thus reassuring the villagers; and (II) the erionite concentrations in the school buses transiting on erionite roads in North Dakota were as high as in the old mesothelioma village. In response to these findings, and upon Carbone's team request, the North Dakota State Authorities repaved all erionite roads, thus averting a possible future mesothelioma epidemic (7). The research in Cappadocia was having an impact in the United States and the US EPA was helping the Turkish Health authorities and vice-versa. For this work in 2008 we received the first AACR-Landon Innovator Award for International Collaboration in Cancer Research.

In September of 2010 we started the last year of the P01 grant: still no gene had been found. Baris became ill and could no longer participate in the research. Moreover, strict regulations enacted in Turkey, prohibited shipping DNA samples abroad due to European Union rules. My own ability to participate in the research had been severely limited by the new Directors of the Department of Chest Diseases at the University of Hacettepe, who had replaced Baris, and did not support mesothelioma research. Carbone noted that in one of the US families that he was studying there was a high incidence of mesothelioma and uveal melanoma, two rare cancers, and that these malignancies shared frequent deletions in the chromosome 3p region. This was one of a few remaining regions that genetic linkage analyses and aCGH had indicated as possibly harboring a mesothelioma gene. He decided, as a last attempt, to focus

all the sequencing efforts on 3p. To accelerate the process, he accepted the help of a collaborator, Dr. Testa. This time "luck" was on our side and the post-doctoral fellows working in Dr. Carbone's and Dr. Testa's laboratories identified germline mutations in all affected members of two unrelated US families, while non-affected members in the same families did not carry this mutation (36). These findings were soon confirmed and expanded by many other research teams [reviewed in (37-39)]. Mechanistically, it was found that *BAP1* regulates cell proliferation and DNA repair (40) and that germline heterozygous *BAP1* mutations cause primarily melanomas and mesothelioma (37,41). Moreover, in mice, it was found that heterozygous germline *BAP1* mutations increase the susceptibility to very low doses of asbestos that normally do not cause mesothelioma, supporting Carbone's original hypothesis of gene X environment interaction (42). Mesothelioma are often polyclonal malignancies (43) and in carriers of germline *BAP1* mutations, multiple clones can often grow into separate malignancies in both pleura and peritoneum, a process accelerated by asbestos exposure (44,45).

Because heterozygous *BAP1* mutations cause cancer mostly in people in their 40s and 50s, they do not interfere with procreation, and thus are passed over the course of multiple generations (46). Starting from a few *BAP1* mutant carriers, and using a combination of genealogy and genetics, it is possible to build large family trees and identify additional carriers of germline mutations that can be screened yearly for early detection of melanomas (curable when detected at an early stage). They then can be enrolled in biomarker research trials for detection of mesothelioma. Several promising biomarkers for mesothelioma are currently being tested (47,48). Also chemoprevention is being investigated (49).

Epidemiological studies revealed that carriers of germline *BAP1* mutations experience prolonged survival following the development of mesothelioma and other malignancies, a finding that may be in part related to early detection as these families are closely monitored for early signs of malignancy (3,11). Unfortunately, this finding does not apply to mesothelioma patients in Cappadocia, possibly because a gene other than *BAP1* predisposes Cappadocia families to mesothelioma, or because early detection strategies are not implemented in Cappadocia. The gene responsible for the high incidence of mesothelioma in some families in Cappadocia has not been identified yet, because it is no longer possible to send DNA specimens from Turkey to the USA. Carbone (3) found that not all US and European families with high incidence of mesothelioma carry germline *BAP1* mutations, suggesting that there are additional genes that when mutated in the germline may increase susceptibility to asbestos/erionite and cause mesothelioma. Potential novel candidate mesothelioma genes were recently identified (35). Moreover, an epidemic of peritoneal mesothelioma among young women in China may provide clues to additional genetic factors that modulate peritoneal versus pleural mesothelioma in women (50). Intriguingly, in Tuzkoy we also noted an unusual higher prevalence of peritoneal mesothelioma in women.

Our studies opened a new and active research field. Several studies demonstrated that *BAP1* acquired somatic mutations are the most common genetic alterations in sporadic (non-hereditary) mesotheliomas, underscoring the critical role of *BAP1* in the pathogenesis of this tumor (35,51-55). Positive nuclear staining for *BAP1* found in close to 100% of lung cancers and negative staining found in about 70% of mesotheliomas, help pathologists in this difficult differential diagnosis (56). Moreover, negative *BAP1* staining helps differentiate benign reactive from malignant mesothelial cells (57), as well as identify biphasic mesothelioma from tumors associated with a benign stromal reaction (58).

Conclusions

The success of this research was the result of human ingenuity combined with highly sophisticated molecular studies—team-work among physician and scientists from different fields and countries determined to find a solution to the tragedy we had seen in three Cappadocian villages. We wanted to fix a problem. The research was not motivated by grants or publications. Moreover, the researchers did not give up because of the many problems encountered associated with conducting medical research in a remote part of the world. This research started in Turkey and ended up helping families in the US and Europe. This research led to the identification of the first, and so far only, gene that modulates mineral fiber carcinogenesis: BAP1. All this started in Cappadocia, and would not have been possible without "the team approach" because none of the investigators had the knowledge to address all the different components of this research. Dr. Baris understood this and welcomed other scientists to work collaboratively; many declined, some for fear of erionite exposure. After our discovery that erionite rather than asbestos was

causing mesothelioma in Cappadocia, our research was not progressing as we had hoped to find a solution to the problem. When we joined forces with Dr. Carbone we made terrific progress, largely because of the enthusiasm he brought to the team with his collaborative approach, determination, drive, and ability to keep an open mind. He could listen and talk to scientists, physicians, and also to people from different walks of life and religion, and then translate this information into scientific hypothesis that we would test. Most importantly, we used our research findings to convince politicians, in Turkey and in the US, to implement preventive measures that are expected to save many lives from mesothelioma and that have already provided a much better quality of life to many.

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Footnote

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References

1. Baumann F, Ambrosi JP, Carbone M. Asbestos is not just asbestos: an unrecognised health hazard. Lancet Oncol 2013;14:576-8.
2. Wylie AG, Candela PA. Methodologies for determining the sources, characteristics, distribution, and abundance of asbestiform and nonasbestiform amphibole and serpentine in ambient air and water. J Toxicol Environ Health B Crit Rev 2015;18:1-42.
3. Carbone M, Kanodia S, Chao A, et al. Consensus Report of the 2015 Weinman International Conference on Mesothelioma. J Thorac Oncol 2016;11:1246-62.
4. Larson D, Powers A, Ambrosi JP, et al. Investigating palygorskite's role in the development of mesothelioma in southern Nevada: Insights into fiber-induced carcinogenicity. J Toxicol Environ Health B Crit Rev 2016;19:213-30.
5. Baumann F, Carbone M. Environmental risk of mesothelioma in the U.S.: An emerging concern - epidemiological issues. J Toxicol Environ Health B Crit Rev 2016;19:231-49.
6. Baumann F, Buck BJ, Metcalf RV, et al. The presence of asbestos in the natural environment is likely related to mesothelioma in young individuals and women from Southern Nevada. J Thorac Oncol 2015;10:731-7.
7. Carbone M, Baris YI, Bertino P, et al. Erionite exposure in North Dakota and Turkish villages with mesothelioma. Proc Natl Acad Sci USA 2011;108:13618-23.
8. Delgermaa V, Takahashi K, Park EK, et al. Global mesothelioma deaths reported to the World Health Organization between 1994 and 2008. Bull World Health Organ 2011;89:716-24.
9. Goossens D, Buck B. Effects of wind erosion, off-road vehicular activity, atmospheric conditions and the proximity of a metropolitan area on PM10 characteristics in a recreational site. Atmospheric Environ 2011;45:94-107.
10. Goossens D, Buck B, McLaurin B. Contributions to atmospheric dust production of natural and anthropogenic emissions in a recreational area designated for off-road vehicular activity (Nellis Dunes, Nevada, USA). J Arid Environ 2012;78:80-99.
11. Baumann F, Flores E, Napolitano A, et al. Mesothelioma patients with germline BAP1 mutations have 7-fold improved long-term survival. Carcinogenesis 2015;36:76-81.
12. Wagner JC, Sleggs CA, Marchand P. Diffuse pleural mesothelioma and asbestos exposure in the North Western Cape Province. Br J Ind Med 1960;17:260-71.
13. Lanphear BP, Buncher CR. Latent period for malignant mesothelioma of occupational origin. J Occup Med 1992;34:718-21.
14. Linton A, Vardy J, Clarke S, et al. The ticking time-bomb of asbestos: its insidious role in the development of malignant mesothelioma. Crit Rev Oncol Hematol 2012;84:200-12.
15. Henley SJ, Larson TC, Wu M, et al. Mesothelioma incidence in 50 states and the District of Columbia, United States, 2003-2008. Int J Occup Environ Health 2013;19:1-10.
16. Turkey Cancer Statistics 2014. Available online: http://kanser.gov.tr/
17. Artvinli M, Gönen Ö, Barış Yİ, et al. Türkiye'de asbestosis. CTFD 1974;5:438-53.
18. Baris YI. Asbestos and Erionite Related Chest Diseases. Ankara-Turkey 1987:36-102.
19. Selçuk ZT, Cöplü L, Emri S, et al. Malignant pleural mesothelioma due to environmental mineral fiber exposure in Turkey. Analysis of 135 cases. Chest 1992;102:790-6.

20. Karakoca Y, Emri S, Cangir AK, et al. Environmental Pleural Plaquesdue to asbestos and fibrous zeolite in Turkey. Indoor and Built Environ 1997;6:100-5.
21. Emri S, Demir A, Dogan M, et al. Lung diseases due to environmental exposures to erionite and asbestos in Turkey. Toxicol Lett 2002;127:251-7.
22. Dogan AU, Baris YI, Dogan M, et al. Genetic Predisposition to Fiber Carcinogenesis Causes a mesothelioma Epidemic in Turkey. Cancer Res 2006;66:5063-8.
23. Baris YI, Sahin AA, Ozesmi M, et al. An outbreak of pleural mesothelioma and chronic fibrosing pleurisy in the village of Karain/Urgüp in Anatolia. Thorax 1978;33:181-92.
24. Sebastien P, Gaudichet A, Bignon J, et al. Zeolite bodies in human lungs from Turkey. Lab Invest 1981;44:420-5.
25. Saracci R, Simonato L, Baris YI, et al. The age-mortality curve of endemic pleural mesothelioma in Karain, Central Turkey. Br J Cancer 1982;45:147-9.
26. Wagner JC, Skidmore JW, Hill RJ, et al. Erionite exposure and mesotheliomas in rats. Br J Cancer 1985;51:727-30.
27. Carbone M, Emri S, Dogan AU, et al. A mesothelioma epidemic in Cappadocia: scientific developments and unexpected social outcomes. Nat Rev Cancer 2007;7:147-54.
28. Carbone M. Simian virus 40 and human tumors: It is time to study mechanisms. J Cell Biochem 1999;76:189-93.
29. Carbone M, Rizzo P, Pass H. Simian virus 40: the link with human malignant mesothelioma is well established. Anticancer Res 2000;20:875-7.
30. Gazdar AF, Carbone M. Molecular pathogenesis of malignant mesothelioma and its relationship to simian virus 40. Clin Lung Cancer 2003;5:177-81.
31. Karakoca Y, Emri S, Bagci T, et al. Environmentally induced malignant plerua mesothelioma and HLA distribution in Turkey. Int J Tuberc Lung Dis 1998;2:1017-22.
32. Kroczynska B, Cutrone R, Bocchetta M, et al. Crocidolite asbestos and SV40 are co-carcinogens in human mesothelial cells and in causing mesothelioma in hamsters. Proc Natl Acad Sci USA 2006;103:14128-33.
33. Emri, S, Kocagoz T, Olut A, et al. Simian virus 40 is not a cofactor in the pathogenesis of environmentally induced malignant pleural mesothelioma in Turkey. Anticancer Res 2000;20:891-4.
34. Roushdy-Hammady I, Siegel J, Emri S, et al. A genetic-susceptibility factor malignant mesothelioma in the Cappadocian region of Turkey. Lancet 2001;357:444-5.
35. Yoshikawa Y, Emi M, Hashimoto-Tamahoki T, et al. High density array-CGH with targeted NGS unmask multiple non-contiguous minute deletions on chromosome 3p21 in mesothelioma. Proc Natl Acad Sci U S A 2016;113:13432-7.
36. Testa JR, Cheung M, Pei J, et al. Germline BAP1 mutations predispose to malignant mesothelioma. Nature Genet 2011;43:1022-5.
37. Carbone M, Yang H, Pass HI, et al. Bap1 and cancer. Nat Rev Cancer 2013;13:153-9.
38. Bononi A, Napolitano A, Pass HI, et al. Latest developments in our understanding of the pathogenesis of mesothelioma and the design of targeted therapies. Expert Rev Respir Med 2015;9:633-54.
39. Napolitano A, Carbone M. Malignant mesothelioma: Time to translate? Trends In Cancer 2016;2:467-74.
40. Daou S, Hammond-Martel I, Mashtalir N, et al. The BAP1/ASXL2 Histone H2A Deubiquitinase Complex Regulates Cell Proliferation and is Disrupted in Cancer. J Biol Chem 2015;290:28643-63.
41. Carbone M, Ferris LK, Baumann F, et al. BAP1 cancer syndrome: malignant mesothelioma, uveal and cutaneous melanoma, and MBAITs. J Transl Med 2012;10:179.
42. Napolitano A, Pellegrini L, Dey A, et al. Minimal asbestos exposure in germline BAP1 heterozygous mice is associated with deregulated inflammatory response and increased risk of mesothelioma. Oncogene 2016;35:1996-2002.
43. Comertpay S, Pastorino S, Tanji M, et al. Evaluation of clonal origin of malignant mesothelioma. J Transl Med 2014;12:301.
44. Hillegass JM, Shukla A, Lathrop SA, et al. Inflammation precedes the development of human malignant mesotheliomas in a SCID mouse xenograft model. Ann N Y Acad Sci 2010;1203:7-14.
45. Qi F, Okimoto G, Jube S, et al. Continuous exposure to chrysotile asbestos can cause transformation of human mesothelial cells via HMGB1 and TNF-α signaling. Am J Pathol 2013;183:1654-66.
46. Carbone M, Flores EG, Emi M, et al. Combined Genetic and Genealogic Studies Uncover a Large BAP1 Cancer Syndrome Kindred Tracing Back Nine Generations to a Common Ancestor from the 1700s. PLoS Genet 2015;11:e1005633.
47. Napolitano A, Antoine DJ, Pellegrini L, et al. HMGB1 and its hyper-acetylated isoform are sensitive and specific serum biomarkers to detect asbestos exposure and to identify mesothelioma patients. Clin Cancer Res 2016;22:3087-96.
48. Ostroff RM, Mehan MR, Stewart A, et al. Early Detection of Malignant Pleural Mesothelioma in Asbestos-exposed Individuals with a Noninvasive Proteomics-based

Surveillance Tool. PLoS One 2012;7:e46091.
49. Yang H, Pellegrini L, Napolitano A, et al. Aspirin delays mesothelioma growth by inhibiting HMGB1-mediated tumor progression. Cell Death Dis 2015;6:e1786.
50. Mao W, Zhang X, Guo Z, et al. Association of Asbestos Exposure With Malignant Mesothelioma Incidence in Eastern China. JAMA Oncol 2017;3:562-4.
51. Nasu M, Emi M, Pastorino S, et al. High Incidence of Somatic BAP1 Alterations in Sporadic Malignant Mesothelioma. J Thorac Oncol 2015;10:565-76.
52. Guo G, Chmielecki J, Goparaju C, et al. Whole exome sequencing reveals frequent genetic alterations in BAP1, NF2, CDKN2A and CUL1 in malignant pleural mesothelioma. Cancer Res 2015;75:264-9.
53. Lo Iacono M, Monica V, Righi L, et al. Targeted next-generation sequencing of cancer genes in advanced stage malignant pleural mesothelioma: a retrospective study. J Thorac Oncol 2015;10:492-9.
54. Bueno R, Stawiski EW, Goldstein LD, et al. Comprehensive genomic analysis of malignant pleural mesothelioma identifies recurrent mutations, gene fusions and splicing alterations. Nat Genet 2016;48:407-16.
55. Ugurluer G, Chang K, Gamez ME, et al. Genome-based mutational analysis by next generation sequencing in patients with malignant pleural and peritoneal mesothelioma. Anticancer Res 2016;36:2331-8.
56. Carbone M, Shimizu D, Napolitano A, et al. Positive nuclear BAP1 immunostaining helps differentiate non-small cell lung carcinomas from malignant mesothelioma. Oncotarget 2016;7:59314-21.
57. Cigognetti M, Lonardi S, Fisogni S, et al. BAP1 (BRCA1-associated protein 1) is a highly specific marker for differentiating mesothelioma from reactive mesothelial proliferations. Mod Pathol 2015;28:1043-57.
58. McGregor SM, Dunning R, Hyjek E, et al. BAP1 facilitates diagnostic objectivity, classification, and prognostication in malignant pleural mesothelioma. Hum Pathol 2015;46:1670-8.

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