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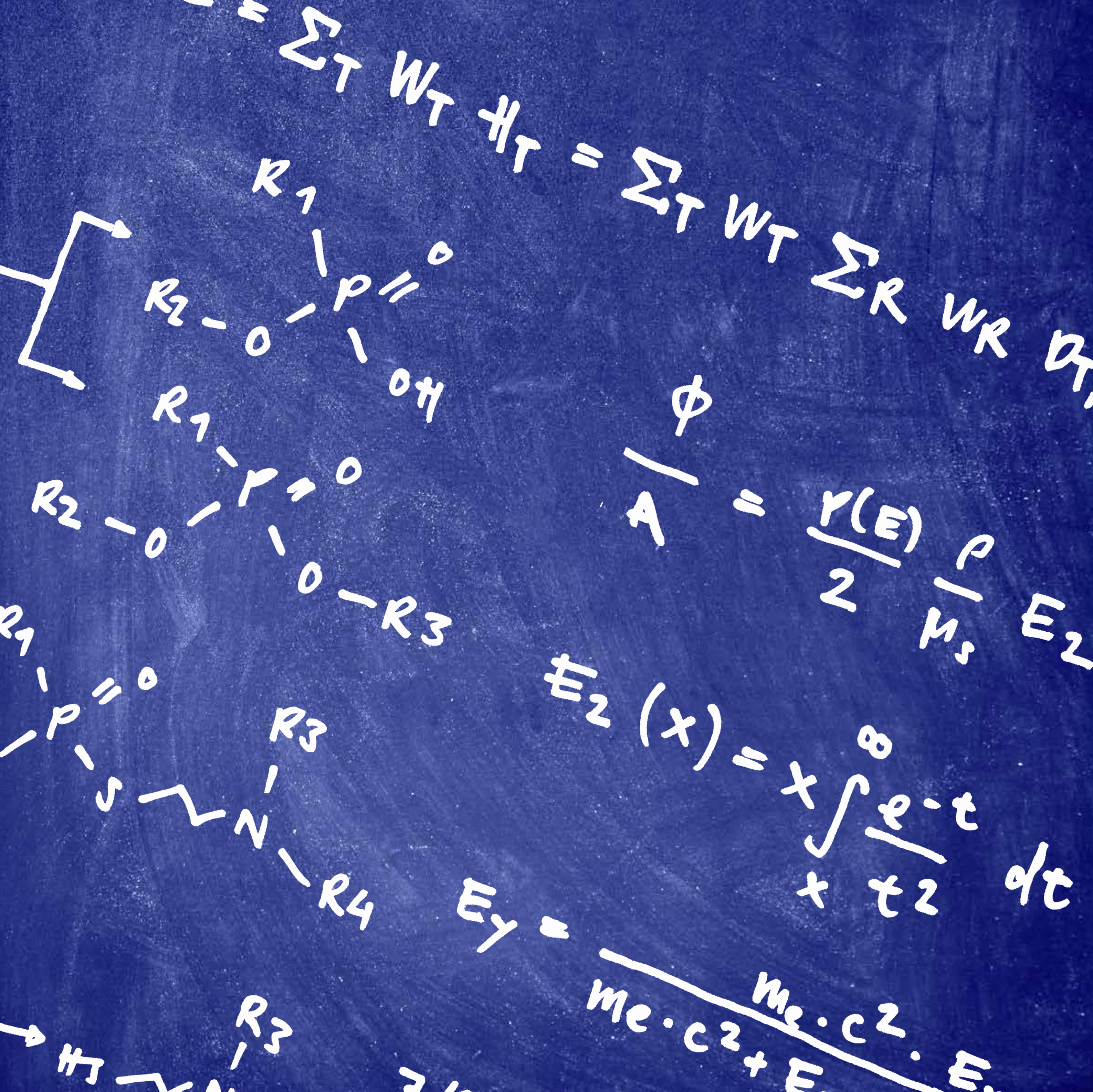
SPIEZ LABORATORY

Annual report 2013

SPIEZ LABORATORY



Production
Dr. Andreas B. Bucher

Design/Layout
Electronic Media Center EMC, 3003 Bern

Editor
Federal Department of Defence, Civil Protection and Sports DDPS,
Federal Office for Civil Protection FOCP
SPIEZ LABORATORY
CH-3700 Spiez
Tel. +41 58 468 14 00
Fax +41 58 468 14 02
laborspiez@babs.admin.ch
www.labor-spiez.ch

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Dear readers,

This report on our activities in 2013 deals primarily with topics that are not yet completed and will keep us busy, not just this year, but probably for much longer.

Above all the crisis in Syria: the use of chemical weapons in the Syrian civil war was verified by the UN mission last autumn. Our Chemistry Division was very heavily involved in this verification task. Last September, it analysed UN/OPCW environmental samples from Syria around the clock for over two weeks and was able to prove without doubt that sarin really had been used in Syria. That we were entrusted with this delicate task underlines our analytical expertise – an expertise that has been consistently confirmed over the years, achieving the highest ratings in inter-laboratory OPCW tests.

In its justification for awarding the Nobel prize to the OPCW, the Nobel prize committee also stated: "Recent events in Syria, where chemical weapons have again been put to use, have underlined the need to enhance the efforts to do away with such weapons." It is precisely these efforts which form part of our core business. In this respect we have, at least from the scientific point of view, contributed to a small extent towards the Nobel Peace Prize awarded to the OPCW. Our work for the OPCW also to an intensive media presence at our research facility in 2013. Our expertise was publicised throughout the world by leading European media such as DER SPIEGEL, the BBC and the Guardian.

This core business of our chemists dominated public interest in the work carried out at Spiez, but our other specialist fields were also successfully involved in arms control and civil protection:

- Amongst other things, the NBC-Protection Division was engaged with quality assurance and the management of individual NBC equipment, including filtering semi-masks as a precaution against pandemics. The results of our quality tests are of considerable interest to both individual households and for security officers employed by commercial firms or serving in the armed forces. (page 34)

- The Physics Division is currently analysing the challenges (decontamination, resettlement, evacuation etc.) faced by the Japanese authorities after the Fukushima incident and identifies what we can learn from Japan that could be relevant for Switzerland. (page 6)

Inspectors of the UN/OPCW joint mission to Syria collect environmental samples near Zamalka/Damascus (August 2013)

Dr. Marc Cadisch
Director SPIEZ LABORATORY

- In the past year, the Biology Division has concentrated on bringing the new biocontainment laboratory into service. The necessary validations have been completed and at the beginning of 2014, the relevant authorities gave their permission for putting it into operation. The next step will be to use the facility for scientifically valuable research projects that are relevant for civil protection: Two projects from the Institute of Microbiology of the University of Lausanne (IMUL) form part of a series of external projects that are to be worked on soon (page 18)

In addition to this work, the development of expertise forms an essential part of our daily work. Our central tasks include keeping track of developments in science, anticipating their impact on civil protection and initiating the necessary measures. In this context we established a chain of international conferences to start in 2014 under the title of "Spiez CONVERGENCE". This workshop is to address the partly overlapping developments in biology and chemistry and indicate possible consequences for the implementation of the chemical and biological weapons conventions.

SPIEZ LABORATORY – as the past has impressively demonstrated – occupies an essential interface between science and politics. Without solid factual knowledge, no progress can be made in disarmament negotiations or in national NBC protection. We are doing our utmost to ensure that we will be able to accomplish this important task in the future too.

The Analytical Chemistry branch with the drafts of the analytical report for the UN/OPCW joint mission in Syria.

Lessons learned from Fukushima

Decontamination and return to normality after an extensive radiological incident

Dr. Emmanuel Egger und Dr. Mario Burger

Since the Fukushima disaster of March 2011, much has been done to make contaminated areas inhabitable again. The prerequisite for a return of the evacuated population is an expected annual dose rate of less than 20 mSv. A return is possible today to relatively few contaminated areas. In more severely contaminated regions, however, radiation exposure still exceeds this limit. This article analyses the situation in Japan towards the end of 2013. It compares the measures taken with those in Belarus after the Chernobyl disaster and we will apply some findings from there to a possible radiological disaster incident in Switzerland.

More than 10,000 km^2 of land were radioactively contaminated by the Fukushima nuclear disaster (Fig. 1). About 159,000 people were evacuated [1]. The Japanese authorities have divided the affected areas into three zones (Fig. 2):

Zone 1: The expected annual dose lies between 1 und 20 mSv. People are allowed to live here or to return if they have been evacuated. Decontamination work is still being carried out. In the long term, the annual exposure dose due to the incident is to be reduced to less than 1 mSv.

Zone 2: The expected annual dose lies between 20 and 50 mSv. People are not allowed to live in this area. Decontamination work is being carried out. It is expected that the annual dose in this area can be reduced to less than 20 mSv in the foreseeable future and that the evacuated population will be able to return. As thousands of square kilometres are affected, the cost will be enormous.

Zone 3: The expected annual dose is higher than 50 mSv. The population has been evacuated. The Japanese authorities now doubt that the population will be able to return to this area in the near future. Although new decontamination methods are being tested, experience to date gives reason to doubt whether the exposure dose can be reduced to less than 20 mSv per annum in the next 2 to 5 years.

Almost three years after the incident, tens of thousands of people still live in makeshift accommodation and do not know whether they will ever be able to return to their former homes. Such a situation could have been avoided if the Japanese authorities had followed the policy of the Soviet authorities after the Chernobyl disaster. However, it is

Government officials check radiation levels of waste from decontamination at a storage site in Fukushima prefecture, 5 July 2013

much more difficult to implement such an approach in densely populated countries like Japan or Switzerland.

Measures after Chernobyl

After the Chernobyl disaster, the Soviet authorities decided to resettle the population of an extensive area. They deliberately dispensed with decontaminating these areas because it is difficult and extremely elaborate to decontaminate entire towns, villages, roads, fields or forests. At the time, the Soviet authorities benefited from the experience they had gained from previous incidents such as that of Mayak in 1957. Experience in Japan confirms that about two man-years are required to decontaminate an area of one hectare. Furthermore, as also confirmed by experience in Japan, the outcome of decontamination is uncertain. The measures taken in Belarus are listed in Table 1 (on page 9).

For the inhabitants affected, these radical measures meant that they quickly knew how things would continue and were thus able to adjust to the new situation.

Recommendations from international organisations

The ICRP (International Commission on Radiological Protection) distinguishes between three special situations concerning radiation exposure [3]:

- Planned radiation exposure = exposure due to deliberate introduction and handling of radioactive sources. Here exposure can be managed and fully controlled.
- Emergency radiation exposure = loss of control over planned exposure (e.g. nuclear power plant incident) or deliberate use of radioactive material (e.g. terror attack).
- Existing radiation exposure = exposure as the result of already existing radioactive sources that should be brought under control.

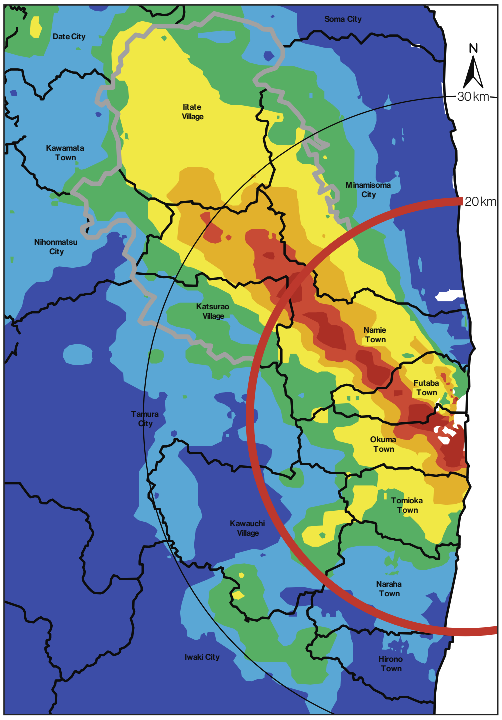

Fig. 1: Expected annual dose in contaminated areas

Measurement by car (mSv/Yr)
150 or more
100–150
50–100
20– 50
10–20
5–10
1–5

Aerial measurement (mSv/Yr)
150 or more
100–150
50–100
20–50
10–20
5–10
1–5

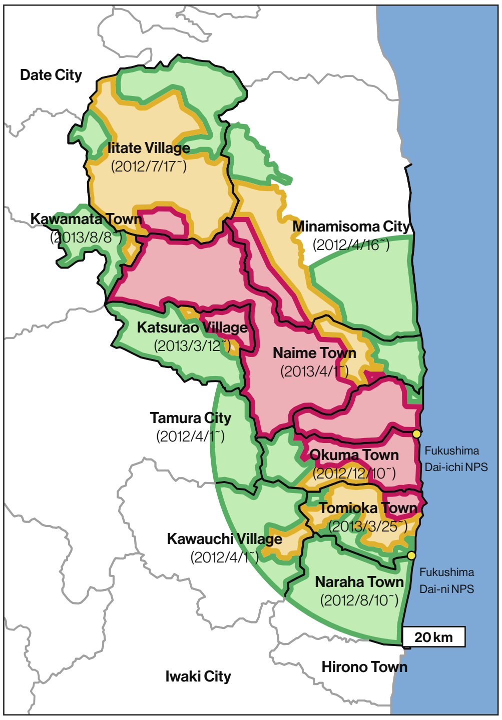

Fig. 2: Contaminated area zones

Area 1: Areas to which evacuation orders are ready to be lifted

Area 2: Areas in which the residents are not permitted to live

Area 3: Areas where it is expected that the residents have difficulties in returning for a long time

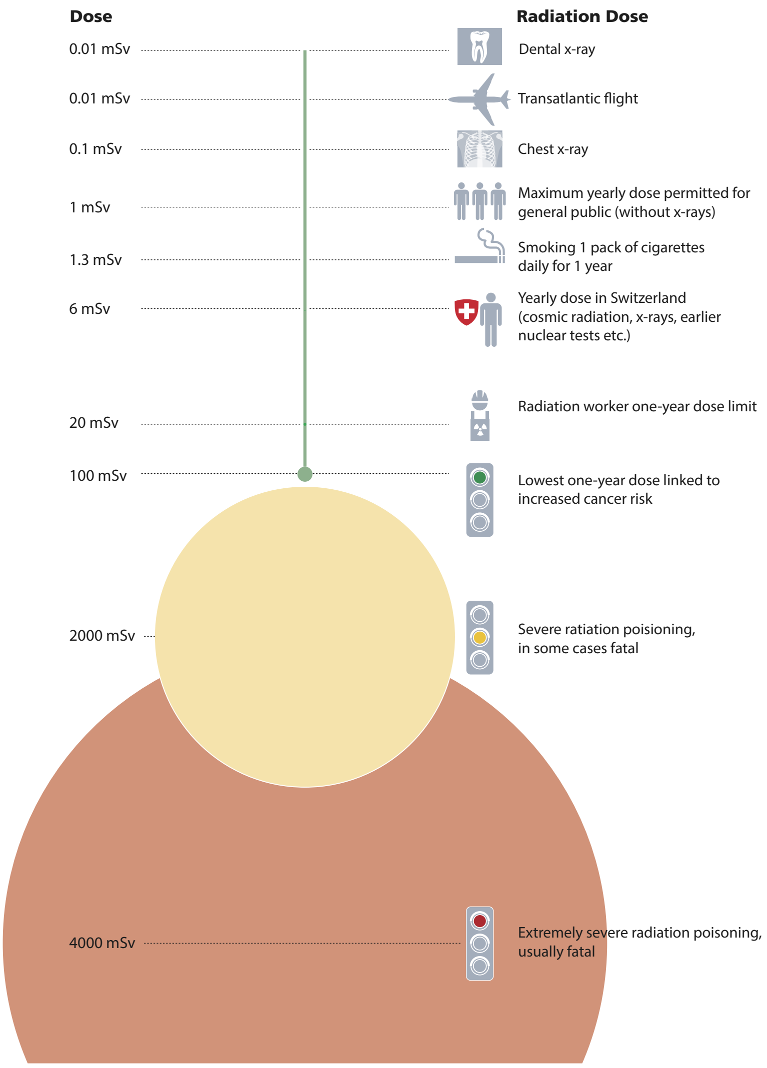

Chart of ionizing radiaton dose a person can absorb from various sources.

The ICRP interprets long-term exposure after an accident like Fukushima as an already existing exposure situation.

To keep radiation exposure as low as possible with reasonable means that take economic and social factors into account, the ICRP recommends the following:

- Radiation exposure should generally be kept under 100 mSv.
- In the event of an incident with possibly long-term consequences, the ICRP recommends:
  - Keeping emergency radiation exposure if possible within 20–100 mSv/year.
  - In the case of existing radiation exposure it should be between 1 and 20 mSv/year, with the long-term goal of reducing it to less than 1 mSv/year.

The transition from emergency to existing radiation exposure is not conclusively defined. It should be defined as soon as sufficient information is available and the situation is under control.

In principle the Japanese authorities pursued exactly this strategy.

These ICRP recommendations, however, do not tell us anything about what measures should be taken to protect the population.

The IAEA recommends taking the following measures [4]:

- If the local dose rate (LDR) exceeds 1,000 μSv/h, the population must be evacuated immediately (within a day) without putting their lives at risk. If the LDR exceeds 25 μSv/h, ten days or more after emission of a radioactive cloud, affected inhabitants should be instructed to prepare for resettlement and to leave the contaminated area within a month. This LDR corresponds to an annual dose outdoors of more than 200 mSv and corresponds to the areas marked in dark red in Fig. 1.

So in Japan it would have been appropriate to resettle the inhabitants of the most severely contaminated areas immediately and not let them hope for three years that they would be able to return to their houses again.

Measure	Cs-137 contamination	Resulting annual
Immediate resettlement	> 1,480 kBq/m²	27 mSv
Resettlement	555 – 1,480 kBq/m²	10 – 27 mSv
Resettlement entitlement	185 – 555 kBq/m²	3.4 – 10.2 mSv
Regular radiation monitoring	37 – 185 kBq/m²	0.7 – 3.4 mSv

Table 1: Measures taken in Belarus following ground contamination after the Chernobyl disaster [2]

Area	Local dose rate LDR prior to decontamination	LDR after decontamination	Reduction of LDR achieved
Residential area	8 µSv/h	4 µSv/h	50 %
Large buildings	9 µSv/h	5 µSv/h	50 %
Fields	11 µSv/h	2 µSv/h	80 %
Streets	9 µSv/h	5 µSv/h	40 %

Table 2: Efficiency of decontamination methods in Japan [6]

In comparison the resettlements ordered after Chernobyl in Belarus went much further than the recommendations of the ICRP.

Limits to decontamination
Decontamination of a radioactively contaminated area means nothing else than removing contaminated material from that area, transporting it away safely and depositing it at a different location. Unlike chemical decontamination, where the chemicals really can be destroyed, this is impossible with radioactive contaminants. Current decontamination methods, as used in Japan too, are pressure washing of roofs and streets, brushing down roofs, etc. Sometimes the road surface is removed and replaced. The top layer of soil is removed from fields. The effectiveness of these methods has been examined and is described in detail in [5]. The conclusion that can be drawn from this, is that the most effective method consists in demolishing buildings and rebuilding them, removing and replacing road surfaces and taking away the top layer of soil from the fields. The disadvantage of these methods is the accumulation of huge amounts of radioactive contaminated material that has to be stored safely.

Methods which preserve the existing structure, such as brushing or vacuum cleaning, have the advantage of producing less waste. Although the water used for hosing down is contaminated, it can be collected and purified. Nonetheless, these methods have the disadvantage that they only remove a fraction of the contaminants. The lessons learned in Japan are listed in Table 2.

In residential areas where the roofs are pressure washed and/or brushed down, a reduction in LDR of approximately 50 % could be achieved. Some streets were pressure washed; in some cases the road surface was removed and replaced. Fields were mainly decontaminated by removing the top layer of soil. That is why decontamination is most effective there.

As already mentioned above, the contaminated areas were divided into three zones. People are allowed to live again in zones with a computed annual dose of less than 20 mSv, whilst further measures are being taken to lower the exposure dose in the long term to less than 1 mSv/year. Areas with an annual dose of 20 to 50 mSv are being decontaminated so that the population can return soon. This should be the case in the foreseeable future. There is however, no expectation that areas with an annual dose rate of more than 50 mSv can be made inhabitable again unless buildings are demolished and disposed of and the top layer of the soil is removed.

Lessons learned from Fukushima
The dose management concept (DMC) of the NBCN Operations Ordinance regulates the measures that would be imposed in Switzerland to protect the population in the event of a radiological incident. Depending on the situation, protected residence (inside a building, in a cellar or shelter) or, if possible, precautionary evacuation would be ordered. If this were impossible, evacuation would be ordered later, after the radioactive cloud has passed by and depending on the level of radiation. Furthermore, for downwind areas, harvesting, grazing, hunting and fishing would be prohibited over a wide area, to ensure that the population did not consume any contaminated foodstuffs. Because such a measure was not imposed in Japan, contaminated food was eaten.

A decontamination worker wipes the tiled roof of a house in Naraha town, which is inside the formerly no-go zone of a 20 km (12 mile) radius around the crippled Fukushima Daiichi nuclear power plant and currently a designated evacuation zone, Fukushima prefecture, 24 August 2013. The most ambitious radiation clean-up ever attempted has proved costly, complex and time-consuming since the Japanese government began it more than two years in the wake of the Fukushima nuclear meltdown. It may also fail. There is also the problem of storage. Most of the contaminated soil and leaves remain piled up in driveways and empty lots because of fierce opposition from local communities to storing it in one place until the Ministry of Environment secures a central site that could hold it for the longer term.

Immediately after the incident, it is of utmost importance to inform the population about the radiation situation. As soon as frightened citizens begin publishing on the Internet individual, unconfirmed readings taken by non-experts, the state has failed. Without exception, measurements must be made according to an established procedure and only with calibrated instruments. If these rules are not observed, the data issued is useless, unwarranted fears are fostered or, in the worst case, a false sense of security prevails.

With its aero-radiometry, Switzerland has a potent resource for rapidly obtaining and communicating the required metric data. Unfortunately Swiss weather conditions only permit the sporadic deployment of helicopters, as was re-confirmed during the integral emergency exercise of November 2013. If weather conditions are bad, Swiss Armed Forces ground radiometry vehicles can be deployed to obtain a picture of the situation. Monitoring troops of the Nuclear DDPS Emergency Team are deployed to obtain more detailed on-site information by means such as measuring the LDR, determining the nuclide distribution through γ spectrometry or sampling for subsequent analysis at SPIEZ LABORATORY.

Although the immediate resources required for dealing with a radiological incident are available in Switzerland, no concept exists for long-term management. Such a concept is currently being developed. It will probably foresee that contaminated areas, similar to Japan, are divided into three zones with different levels of radiation exposure.

In the first zone, the population would be allowed to remain and be temporarily exposed to a slightly higher radiation. The tolerated dose rate still has to be defined. The Japanese authorities set the limit at 20 mSv/year. In the long term, decontamination measures will need to be carried out to reduce radiation exposure due to contamination to less than 1 mSv/year.

In the second zone the population would be evacuated from the area, until it had been decontaminated to less than the tolerated dose rate (in Japan 20 mSv/year). Afterwards, the population would be able to return to the area. In this area too, decontamination would continue, until radiation exposure had been reduced to less than 1 mSv/year.

An extremely extensive monitoring programme would be necessary to optimise decontamination measures in these zones. The contribution made to the local dose rate by every street, roof, tree and garden would have to be determined. It would also be necessary to identify local hot spots. As it is most likely that several weeks or months would pass from the time of the incident until decontamination begins, we can assume that some of the contaminants would have been washed away by precipitation and deposited at the edge of the road or in puddles. Concentrating on decontaminating these hot spots might already considerably lower the LDR. This would present a particular challenge for the contamination monitoring organisation.

After decontamination has been carried out, readings from the area would have to be taken again, to verify its efficacy. Readings would have to be taken in every house, every apartment and every single room, before people were allowed to return to their homes. Here too, a huge effort in terms of time and cost would be likely and could hardly be accomplished with the present monitoring organisation.

In the third zone, radiation exposure would be considered to be too high for the area to be decontaminated to a level below the limit within an acceptable time period. The population living in these zones should definitely be relocated immediately. As in Belarus, a threshold should be defined in which such a measure should be carried out. As many nuclides released after a nuclear power plant incident would have decayed after a few weeks, such a limit would have to be specifically defined to take expected long-lived nuclides (e.g. 1,500 kBq/m^2 for Cs-137) into account. Another approach would be to measure the local dose rate ten days after fall-out and to relocate the population from those areas that exceed a certain local dose rate (e.g. 25 μSv/h). Determining this dose rate would be the task of a working group, assigned to develop a concept for the return to normality. This working group would be able to rely on the recommendations of the ICRP and the IAEA and on previous experience from Japan and Belarus.

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Drinking water analysis in the Democratic Republic of Congo

Alfred Jakob

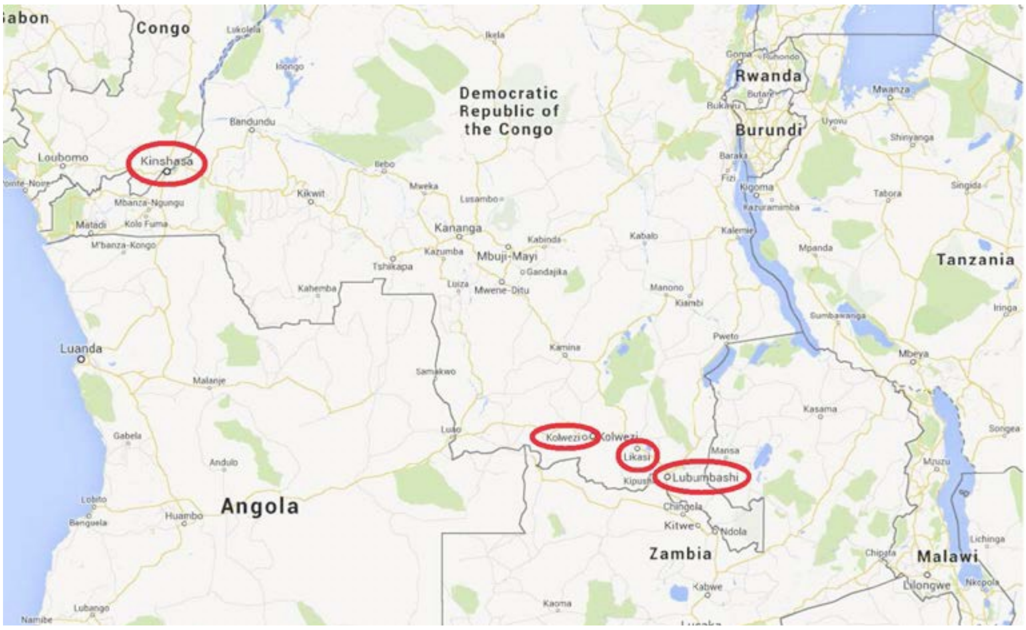
Biologically and chemically polluted drinking water sources are one of the most serious health problems in developing countries. The absence of sanitary installations, broken waste water systems, fertiliser in agriculture, industrial waste water and contaminated soils pollute the ground water and endanger the population. In 2010, more than 35 million people in the Democratic Republic of Congo had no access to clean drinking water. As commissioned by UNEP and UNICEF, SPIEZ LABORATORY supported a "Water Safety Plan" (WSP) project in 2013 as part of the major "VEA – Villages et Ecoles Assainis" development project.

The national VEA (Villages et Ecoles Assainis) project was started in 2006 and has since then been supported by UNICEF. The overriding goal is to provide four million people in about 6,000 villages and 1,250 schools with clean drinking water by 2017. In the Congo drinking water is severely polluted by domestic waste water (excrements) with pathogenic microorganisms. Waste water from mines can contain acids and heavy metals that jeopardise the ecology of rivers and threaten fish populations. In the major mining areas that particularly affect the rivers in the south of the Democratic Republic of Congo, ground water may also be contaminated today with toxic heavy metals that are hazardous to health.

Safe drinking water is clear, colourless, odourless, tastes good and is neither contaminated by microbes nor by chemicals. To meet these requirements, ground water and its reservoirs must be protected against pollutants.

By the end of 2012, it was possible to improve hygienic conditions in about 3,000 villages and 1,000 schools in the country thanks to education and sanitary installations provided in this project.

To evaluate the efficiency of measures taken to date and to find out what further measures might be required, SPIEZ LABORATORY examined about 50 drinking water cisterns in select rural and urban regions of the country. The mobile laboratory of the Swiss Humanitarian Aid corps (SHA) was used to determine the microbiological and chemical quality of drinking water. Field measuring equipment enabled



Analysis of drinking water inlets in selected rural and urban areas of the Democratic Republic of Congo

examination of turbidity, conductivity, colour, smell, temperature and acidity of water samples and the analysis of various chemical parameters, as well as a very informative on-site microbiological test for each drinking water source. The detailed chemical analyses of samples to comprehensively determine drinking water quality were made by the Environmental Analysis Branch in Spiez.

Field measurements at a water source

The trace concentrations (micrograms per litre) of toxic heavy metals such as cadmium, arsenic, mercury and cobalt were determined by the elementary ICP-MS analysis method (inductively coupled plasma mass spectrometry). The analysis of mineral salt concentrations in drinking water and the concentrations of the pollutant indicators nitrate and ammonia were established through ion chromatography (IC).

Analysis of water samples in Spiez

Numerous talks with health authorities and universities were held in the course of the project. Local laboratories were visited to assess their analytical competencies for evaluating drinking water and to determine the measures required to build the necessary analytical infrastructure (laboratory capacity building).

The analytical results of the mission in 2013 and derived project findings enabled both mandate givers, the UNICEF and the Congolese health authorities, to introduce substantial steps to further improve the quality of drinking water and thus the population’s quality of life.

(http://www.ecole-village-assainis.cd/)

Radio nuclides in Lake of Bienne sediments

Dr. Stefan Röllin

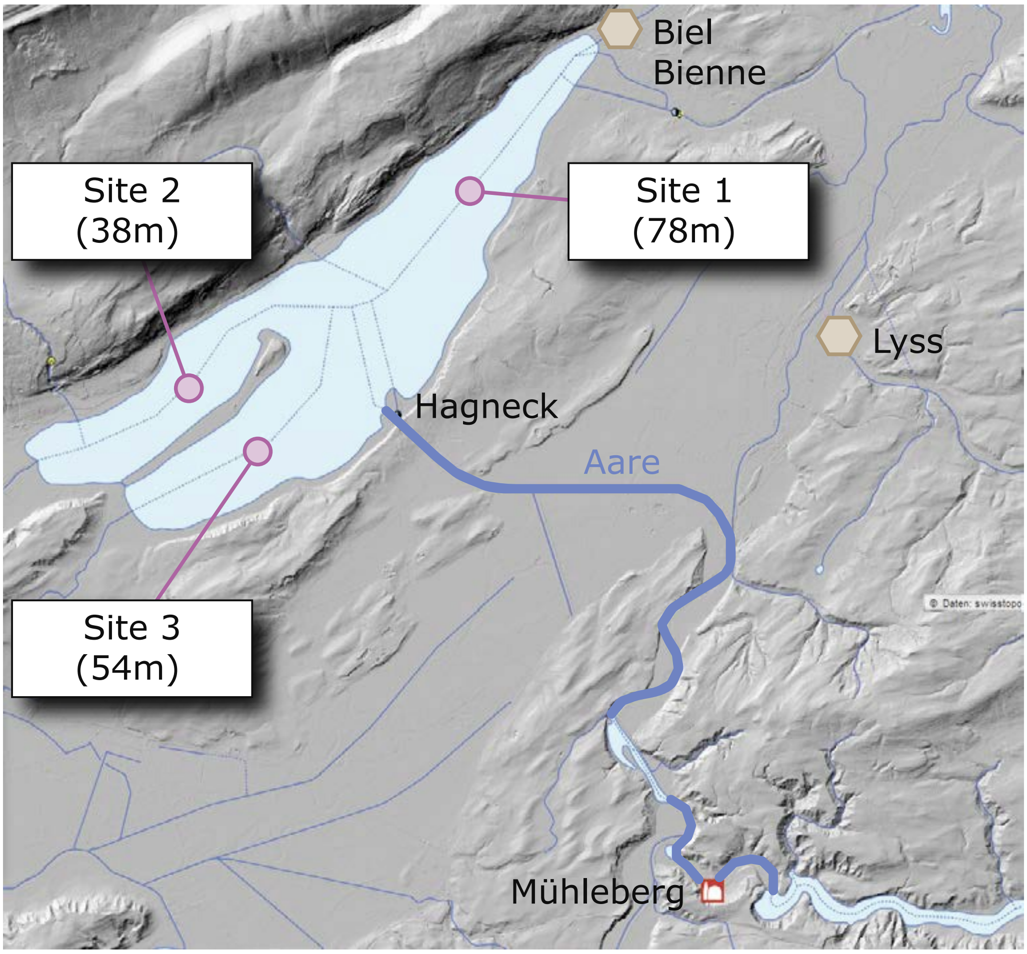
Within the context of radioactivity monitoring in Switzerland, the Radioactivity Branch has been monitoring sediment profiles from various lakes in cooperation with the University of Ravensburg-Weingarten since 1995. Sampling from the Lake of Bienne planned for 2013 attracted great interest as shortly before Swiss media had reported in accordance with the publication of Thevenon (Thevenon 2013) that a slightly higher input of radioactive Cs-137 had been deposited in the Lake of Bienne around 2000.

The history of a lake can be reconstructed from sediment cores: Every year, a fresh sediment layer settles to the bottom of the lake. The individual strata show up very well in a depth profile. A relationship between age and depth can be deducted from the two clearly discernable Cs-137 rich deposits from the Chernobyl nuclear power plant incident in 1986 and signatures due to nuclear weapon tests with the most active one being in 1963. Already in 1998, the Swiss Federal Institute of Aquatic Science and Technology (EAWAG) was able to show that during the interval between the typical Cs-137 signatures of 1986 and the 1960s there had been other clearly discernable deposits of Cs-137 in the Lake of Bienne caused by material released from the Muhleberg nuclear power plant. In addition, there was also evidence of Co-60 that had come from the Muhleberg nuclear power plant.

The aim of the work in 2013 presented here, was to verify higher levels of Cs-137 deposited around 2000 and to obtain indications of their origin.

Sampling and processing

The Lake of Bienne consists of two basins that lie to the south-west and the north-east of a narrow peninsula and unite beyond the peninsula to form a further third common basin. The body of water to the north of the peninsula is fed by the Zihl canal with water from the Lake of Neuchatel, while the southern basin is fed by the Aare or the Aare-Hagneck canal. Water exits the common basin of the Lake of Bienne via the Nidau-Büren canal and follows the original riverbed of the Aare. The Muhleberg nuclear power plant, a pressure water reactor that has been in service since 1971, is located on the bank of the Aare River and uses its water for cooling. Sediment cores BL13-1a,



Sediment cores BL13-1a, BL13-2d and BL13-3d were taken from the deepest points of the three Lake of Bienne basins on 17 and 18 July 2013. The cores were analysed by the aquatic research institute EAWAG and SPIEZ LABORATORY, in cooperation with the University of Ravensburg-Weingarten and the Institute of Geography of the University of Berne.

BL13-2d and BL13-3d were taken from the deepest points of the three Lake of Bienne basins on 17 and 18 July 2013 respectively.

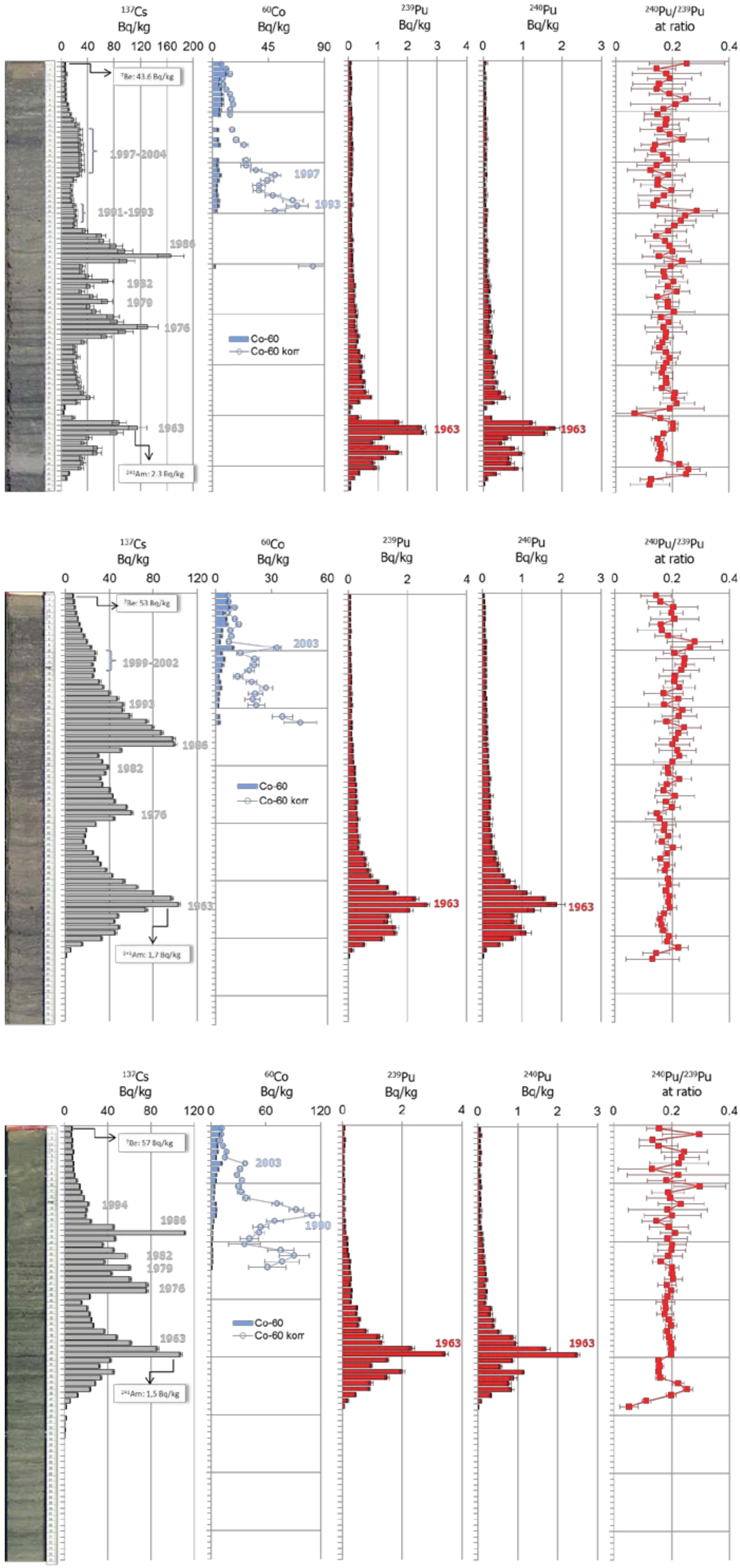
Sampling was carried out in cooperation with the EAWAG (Fig. 2). The sediment scoop was equipped with an interchangeable plastic pipe with an inner radius of 6 cm and was driven into the sediment by its own weight. Thus obtained sediment cores of approximately 1 meter length were halved lengthwise, photographed and divided into 1 cm thick layers. The samples were weighed in polystyrene containers, freeze dried and subsequently homogenised and then each measured for a day by gamma spectrometry (Cs-137, Co-60). The samples were incinerated and their components extracted by melting for mass spectrometry (Pu-239, Pu-240). For analysis of plutonium, plutonium was also separated by means of extraction chromatography.

The average sedimentation rates were determined for the 2013 – 1986 and the 1986 – 1963 periods to determine the relationship between depth and age:

Site	Time	R in cm/year
BL13-1a	2013–1986	1.41
	1986–1963	1.48
BL13-2d	2013–1986	0.96
	1986–1963	1.22
BL13-3d	2013–1986	0.67
	1986–1963	0.91

Average sedimentation rates (R) at the three sites in the Lake of Bienne

The vertical distribution of Cs-137 reveals clear increases between 1972 and 1986. These deposits correspond well with the releases reported by the nuclear power plant operators. After 1986, it was also possible to detect slight increases in Cs-137 in the sediments. In two sediments it was possible to confirm a slight increase around 2000. Additional cores were taken from all three locations and analysed by the EAWAG. The Cs-137 activities in cores taken from the same locations corresponded very well. In the case of the additional cores, however, a slight increase in Cs-137 was found for all locations in 2000. Such differences could be explained by the displacement of sediments. If we take such uncertainties into account, these deposits agree with the increased releases declared by the Muhleberg nuclear power plant for 1999. The Cs-137



Vertical distribution of non-natural radio nuclides in the sediment cores of the three locations BL13-1a, BL13-2d and BL-13-3d in the Lake of Bienne.

The photo of the lengthwise halved sediments can be seen on the left. Seasonal light-dark stratification is clearly evident in all sediments. Some of the strata can also be explained by individual incidents such as storms.

In the diagrams to the right of the photographs the respective activity concentrations per dry weight are listed. Signatures resulting from the Chernobyl reactor incident in 1986, and the nuclear weapons tests of the 60s as well as the exponentially declining signature from the source area of the lake are clearly evident in the depth profile of Cs-137.

values measured constitute no health hazard and are well below the safety limits as defined by the Federal Department of Home Affairs FDHA in the Xenobiotic Substances and Components Ordinance. Other than in sediments taken from the lakes of Thun and Brienz (Röllin 2011), it is possible to detect Co-60 in the sediments of the Lake of Bienne. Due to the relatively short half-life of Co-60 (5.3 years) radioactivity levels decline exponentially with depth. But even decay-corrected Co-60 activity concentrations (Co-60 corr), clearly show that the Co-60 signature from releases by the Muhleberg nuclear power plant has become weaker over the years.

The vertical distribution of plutonium isotopes reveals deposits due to nuclear weapon tests only. This is confirmed by the isotope ratio of Pu-240/Pu-239. A value of 0.18 is typical for plutonium from global fallout. A higher isotope ratio would be expected for plutonium from the Muhleberg reactor. Whether the slightly higher isotope ratio of plutonium could have been caused by releases from the Muhleberg nuclear power plant, would need to be verified with more precise isotope ratio measurements.

Sampling from the Lake of Bienne in 2013 (colleagues from the University of Ravensburg-Weingarten and the EAWAG)

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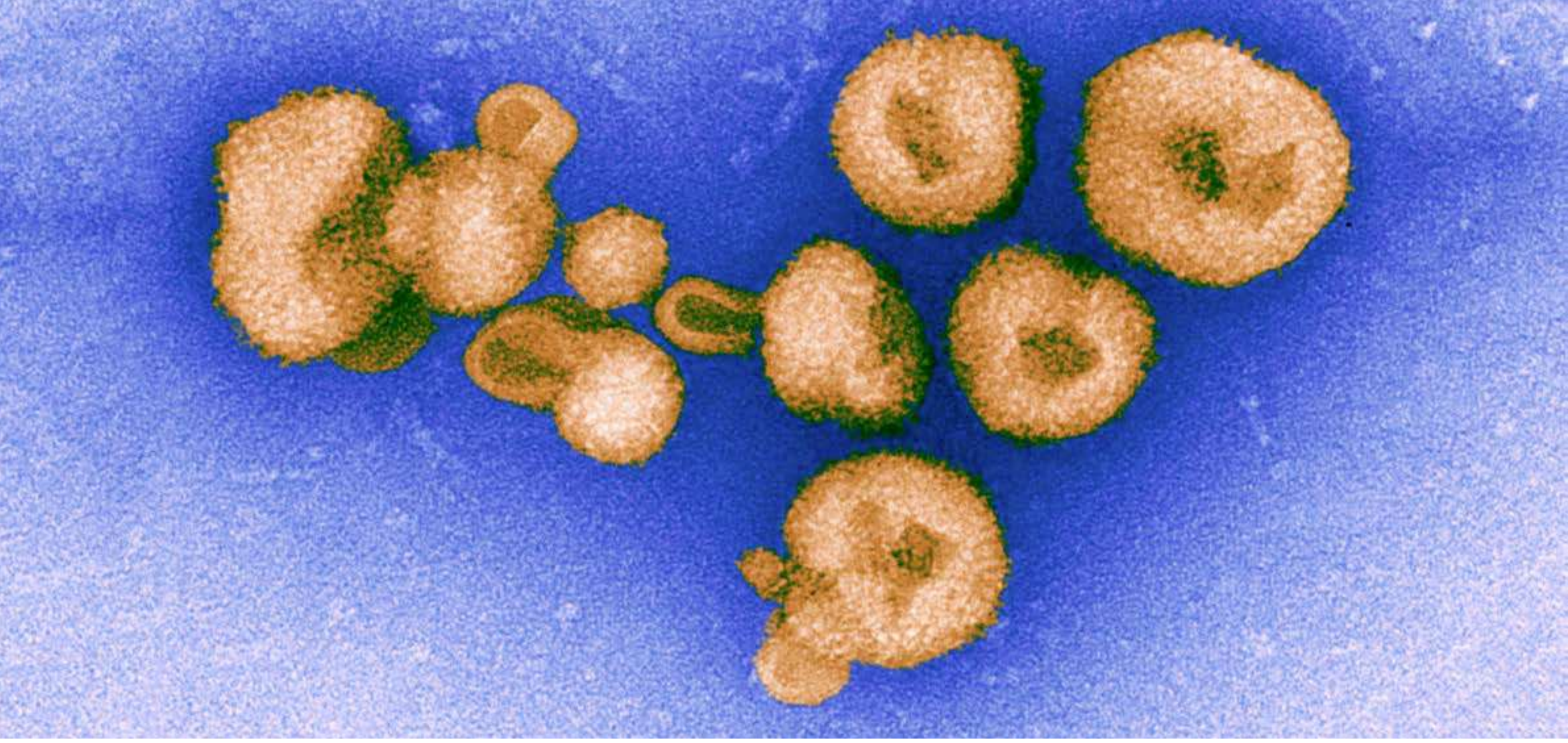
See also: FOPH, Abteilung Strahlenschutz Umweltradioaktivität und Strahlendosen in der Schweiz 2013, Radionuklide in Sedimenten des Bielersees

S. Röllin, Labor Spiez, Bundesamt für Bevölkerungsschutz des VBS, 3700 Spiez;
J. Beer, Eawag, Abteilung Surf/RT, 8600 Dübendorf;
B. Balsiger, Labor Spiez, Bundesamt für Bevölkerungsschutz des VBS, 3700 Spiez;
M. Brennwald, Eawag, Abteilung W+T, 8600 Dübendorf;
S. Estier, Sektion Umweltradioaktivität URA, BAG, 3003 Bern ;
E. Klemt, Hochschule Ravensburg-Weingarten, D-88250 Weingarten;
A. Lück, Eawag, Abteilung Surf/RT, 8600 Dübendorf;
V. Putyrskaya, Hochschule Ravensburg-Weingarten, D-88250 Weingarten;
H. Sahli1, Labor Spiez, Bundesamt für Bevölkerungsschutz des VBS, 3700 Spiez;
P. Steinmann, Sektion Umweltradioaktivität URA, BAG, 3003 Bern



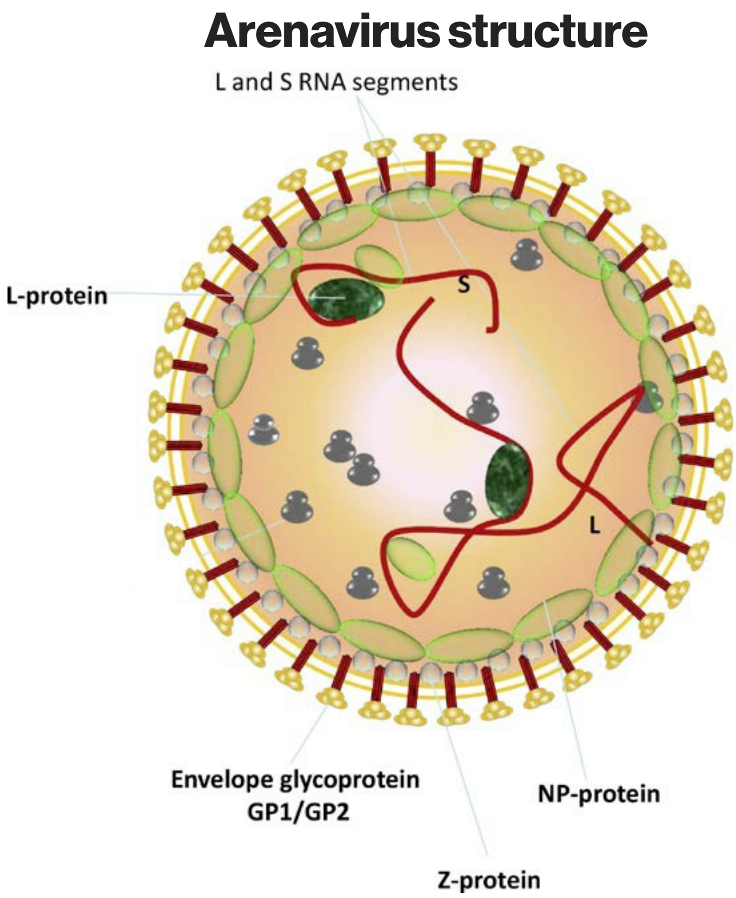
Research projects in the Biocontainment Laboratory

Prof. Dr. Stefan Kunz, Prof. Dr. Stephen Leib

In 2014, the highest safety level of the biocontainment laboratory at SPIEZ LABORATORY will become operational. The work planned for the laboratory will serve to diagnose highly dangerous pathogens and to research the spread and pathogenesis of diseases and therapeutic options. The projects must be relevant to civil protection, have to be based on scientific principles and must address scientifically and socially important issues and provide reliable answers.

Two collaborative projects with the Institute of Microbiology of the University of Lausanne (IMUL) form part of a series of projects that are to be worked on after the biocontainment laboratory in Spiez has become operational; one important reason being that only SPIEZ LABORATORY has the necessary safety facilities for carrying out these studies. Both projects employ new and promising technologies to solve a highly relevant biomedical problem and could thus prepare the way for developing a first vaccine against haemorrhagic Arena viruses. Over the past decades, several Arena viruses have often emerged in humans as pathogens giving rise to the frequently deadly haemorrhagic fever (VHF). In Africa the Lassa virus infects hundreds of thousands of patients every year and currently threatens more than 180 million people. On the American continent the South American haemorrhagic fever viruses Junin, Machupo, Guanarito und Sabia are responsible for local epidemics with a high mortality rate. International air traffic regularly imports Arena virus VHF cases to major cities all over the world which threaten local inhabitants. Because of their potential danger, haemorrhagic fever viruses have been classified by the Centres of Disease Control and Prevention as Category A agents, since they pose a considerable danger to public health and safety. There is currently no generally licensed vaccine against Arena viruses available and current therapeutic means are very limited. The development of new effective medicinal products and vaccinations against these deadly viruses is therefore a high priority.

A main goal of the research programme is identifying host cell factors that pathogenic Arena viruses require for their propagation. In the applied part of the programme, these essential cellular factors will be evaluated as therapeutic targets for antiviral medication.



Over the past decades several arenaviruses have emerged as causative agents of severe viral hemorrhagic fevers (VHF) with high mortality in man.

An interdisciplinary project with Prof. Melody Swartz from the École polytechnique fédérale de Lausanne (EFPL) is studying the development of a vaccine against Arena human pathogen viruses, based on a new nanoparticle platform.

Project 1: Development of new medicinal products against Arena virus pathogens
In the case of haemorrhagic fever caused by Arena viruses, the lethal course of the disease is closely correlated to the viral load, suggesting a competition between viral propagation and the patient's immune response. Therapeutic agents that block various steps in the viral propagation cycle could slow down viral propagation, providing the patient's immune system with the time required to develop an efficient immune response.

The development of antiviral drugs is currently limited through the rather incomplete knowledge of the molecular structure of Arena viruses. However, as obligatory cell parasites, Arena viruses are critically dependent on the molecular machinery of the host cell for their propagation. Therefore, host cell factors necessary for viral replication, provide interesting therapeutic targets for antiviral medication. The applied research branch of IMUL identifies such essential host cell factors and develops specific inhibitors which are then evaluated as candidates for antiviral medication; for example, the cellular proprotein convertase subtilisin kexin isozyme 1 (SKI-1)/site 1 protease (S1P) that is crucial for the biosynthesis of the fusion-competent viral glycoprotein (GP) of the viral envelope. Studies to date have shown that this molecular mechanism is essential for infection and dissemination of Arena viruses [1–6]. In experiments with human cell cultures it has been possible to confirm a strong antiviral effect of SKI-1/S1P inhibitors on the propagation of human pathogen Arena viruses [7–10]. In order to evaluate whether these SKI/SPI inhibitors can be used for antiviral therapy, they must also be examined in a complex living organism. For this purpose it is planned to use a new mouse model to study infection with the Lassa virus [11] and the Machupo virus [12]. In cooperation with SPIEZ LABORATORY, the prophylactic and therapeutic potential of SKI-1/S1P inhibitors as antiviral therapeutics is to be tested in vivo in the biosafety lab.

Support: SNF Grant FN 310030-149746
SNF Grant FN 31003A-135536
University of Lausanne

Project 2: Development of an Arena virus vaccination based on nanoparticles
Due to the currently inadequate therapeutic options, the development of a safe vaccine against pathogenic Arena viruses has a high medical priority. Within the context of an interdisciplinary project between IMUL and the research groups from the Institute of Bioengineering of the EPFL a new recombinant vaccine against human pathogenic Arena viruses was developed. During the first phase of the project, a vaccine was developed that is based on a nanoparticle formula that is able to initiate a T-cell response. Creating vaccine formulas, however, that are able to trigger the production of what are called neutralising antibodies against human pathogenic Arena viruses presents a particular challenge. For this purpose, a new type of nanoparticle was developed that presents the viral protein (antigen) in its native conformation which greatly enhances the chances of eliciting a neutralising antibody response. Studies in mice that can only be carried out in the biocontainment lab should allow investigation into whether a protective effect can be achieved with this newly developed vaccine.

Support: SNF Interdisciplinary Grant FN CR2312-143754

Limited and well justified experiments in mice and rats are an indispensable element in developing new therapy options. Animal studies are of essential importance for demonstrating and further perfecting the immunological procedures of vaccine protection. Thus, the researchers at IMUL are only able to test candidates for nanoparticle vaccines against Lassa und Machupo viruses thanks to animal experiments. The Biology Division of SPIEZ LABORATORY will conduct animal studies in the new biocontainment lab in close collaboration with university partners. The animal experiments are planned so that the number of animals and in particular the degree of distress on the animals can be kept as low as possible. Precise criteria for termination of the experiments have been defined and the animals will be monitored on a daily basis. To achieve this, an animal holding and experiment unit will be established at the biocontainment lab in accordance with the Federal Veterinary Office's Ordinance of 12 April 2010 on Keeping Laboratory Animals und Animal Experiment Procedures. Contractual support by Dr. med. vet. Max Müller, head of the Experimental Animal Centre of the Medical Faculty of the University of Bern, will enable us to set up the appropriate infrastructure for keeping laboratory animals.

Switzerland has one of the world's most comprehensive animal protection legislation systems with particularly stringent regulations for experiments on animals:

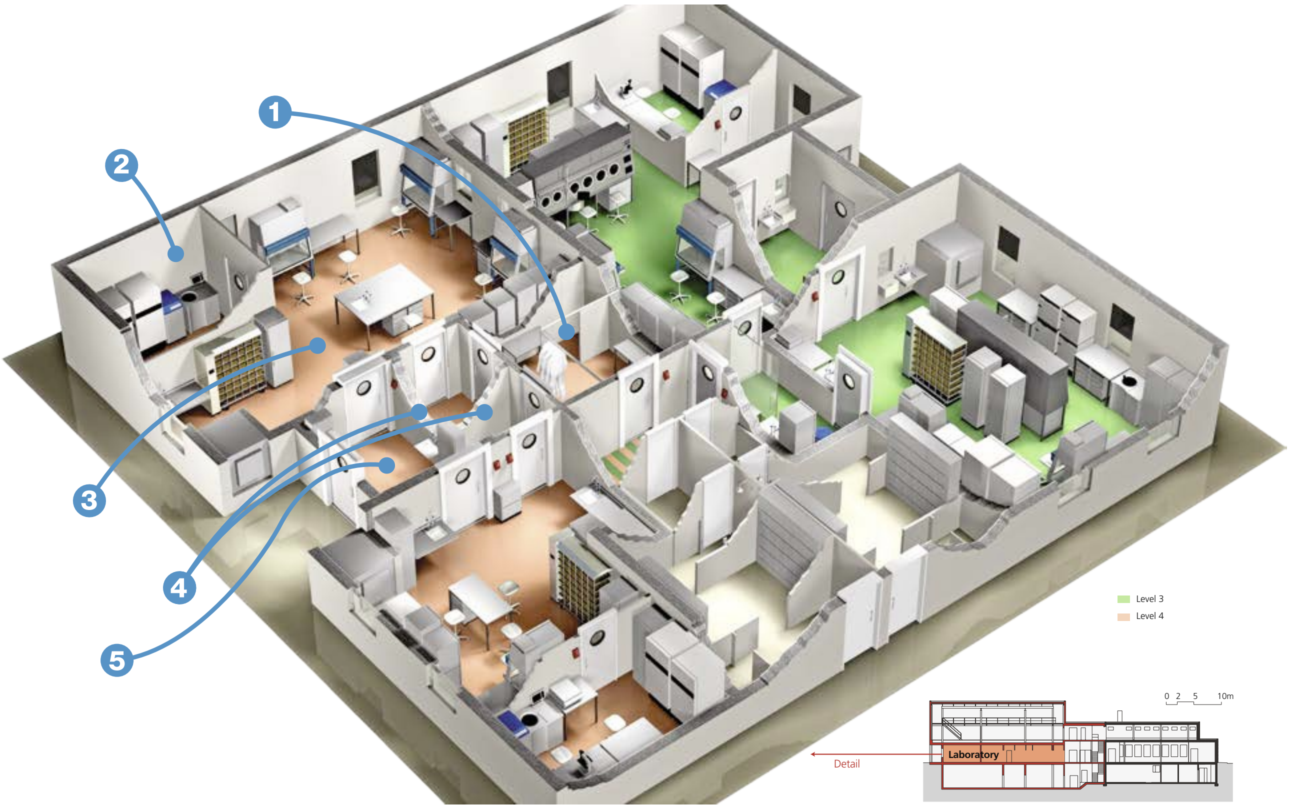
- In Switzerland each animal experiment has to be justified and authorised. Various agencies are involved under a mutual monitoring regime. An important role is played by animal experiment committees on which animal protection organisations are also represented.

- The researchers are obliged to show that the benefit of the experiments for society is greater than the suffering of the animals (balancing of competing interests) and that no alternative methods exist.

- The Animal Care and Experimentation Comitee of the Canton of Berne reviews the applications and issues a recommendation: refusal, permission under certain conditions (e.g. special method, number of animals, species) or general permission.

- Whoever conducts animal experiments must have the necessary expertise. Since 1999, researchers must have been trained in animal protection and must regularly attend advanced training sessions. Staff members who care for the laboratory animals must also have received appropriate training.

Set-up of the biocontainment laboratory in Spiez



The Biosafety-Level 4 in Spiez

1. Suit Room
2. Dark Space with Ultra Centrifuge and Fluorescence Microscope
3. Incubators, freezers, biosafety cabinets, double-door autoclave, animal holding and experimentation unit
4. Chemical showers
5. Material air lock

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Ricin in bio fertiliser?

Marc-André Avondet

Ricin is a toxic protein that is present in the seeds of the ornamental and crop plant Ricinus communis. Due to its potency, and because large amounts are potentially available, this toxin is regulated as a Schedule 1 compound in the Chemical Weapons Convention (CWC). As it arises as a by-product in industrial castor oil production, there is also a certain risk that deliberate or accidental poisoning may occur with manufacturing residues. Since 2000, there have been several cases involving biological fertilisers in Germany. In 2013, SPIEZ LABORATORY examined some similarly suspect samples from Switzerland.

At the end of May 2013, a request was received by SPIEZ LABORATORY from a dog owner living in eastern Switzerland. He asked for assistance in clarifying the death of his four-legged friend in March 2013. The owner believed that his dog had died of ricin poisoning [1, 2], caused by the consumption of biological fertiliser containing ricin and ricinine. This suspicion was published in an article in the consumer magazine K-Tipp of 22 May 2013 [3]. No evidence was collected nor were there any definite findings to indicate the exact cause of the animal's death. Consequently, SPIEZ LABORATORY was only able to provide assistance in the form of information or advice. This was the first such case involving biological fertiliser in Switzerland.

The dog owner approached the Federal Office for Agriculture with the argument that a biological product containing a highly poisonous substance should be prohibited. The FOA agreed and in the middle of June 2013, issued an immediate prohibition on using additives that could contain ricin (Ricinus communis shreddings) in fertiliser production [4]. Oekohum, the Thurgau-based manufacturer of biological fertiliser, did not agree with this decision and demanded an interim period to enable it to run down its existing stocks. So in order to clarify the facts, the company commissioned SPIEZ LABORATORY at the beginning of June to determine the ricin content of its Ricinus communis shreddings and biological fertiliser. This request was accepted with the agreement of the FOA. The head of its biological fertiliser department welcomed an investigation that would enable a correct assessment to be made of the extent of the problem. At the time, the Oekohum company was faced with high compensation demands from the affected dog owner.

Since 1980, there have repeatedly been cases of dog poisoning, especially in Germany, in conjunction with what is known as biological fertiliser. These biological fertilisers usually consisted of a mixture of by-products from the animal industry (blood, bone or keratin powder) as well as press residues from castor oil production, Ricinus communis shreddings. The addition of these by-products make biological fertiliser attractive for dogs, so that the animals could quite possibly eat fairly large amounts.

Because deactivation (heat treatment) of the ricin contained in press residues had either not been carried out or was incomplete, the dogs were poisoned and quite a few died as a consequence. In 2010, there was another wave of poisonings, the cause again being biological fertiliser that was relatively rich in ricin and ricinine [5]. Nine dogs were poisoned and the pathological analysis revealed the typical symptoms of severe haemorrhagic gastroenteritis. Lab analyses produced a figure of 1.715 mg/kg ricin in biological fertiliser, the appropriate limit being 50 mg/kg (EU) [6].

Compared with the biological fertilisers sold in Germany, the Swiss biological fertiliser was based on a purely vegetable formula consisting of rapeseed and Ricinus communis shreddings without animal additives. This begs the question of the extent to which dogs are at all interested in a vegetarian product. As no current samples could be examined and no pathological study was made of the dog either, it has not been proven that ricin from the biological fertiliser really did cause the dog's death.

Examination of the Swiss biological fertiliser
Representative random samples were taken from the three specimens that were provided in large amounts (several kilograms) and subsequently ground to ensure homogeneity for analysis. In a further step the samples were extracted using a method developed by the Robert Koch Institute in Berlin. This method extracts water soluble components (including ricin) from the samples with a PBS buffer; the samples are then centrifuged and filtered.

These extracts were used for determining the ricin concentration through an immunological detection procedure (ELISA) [7]. A methanol extract was used for determining ricinine concentration by chromatography (HPLC-DAD).

Sample 1 = Ricinus communis shreddings (original condition or ground)

Sample 2 = Ricinus communis shreddings (pelleted or ground)

Sample 3 = biological fertiliser (pelleted or ground)

The manufacturer of the fertiliser provided SPIEZ LABORATORY with three different samples for analysis. The first sample was Ricinus communis shreddings as it arises as a by-product of castor oil production. The second sample consisted of shreddings that had been extruded to form pellets. The extrusion was carried out at temperatures of approx. +80°C. The third sample was the pelleted biological fertiliser consisting of rapeseed and Ricinus communis shreddings.

Table 1: Results of ricin and ricinine measurement [8]

Samples	Ricin concentration [mg/kg] (n=4)	Ricinine concentration [mg/kg] (n=2)
Ricinus communis shreddings (Sample 1)	810	1,700
Pelleted Ricinus communis shreddings (Sample 2)	170	2,100
Pelleted biological fertiliser ((Sample 3)	120	840

Table 1 shows the results of the analysed samples. Their ricin content was also determined by the Microbiological Toxins Group at the Robert Koch Institute in Berlin. The results corresponded well with those from the Toxinology Branch of SPIEZ LABORATORY. The ricin bioactivity of the samples was determined by functional assay (cytotoxicity assay with Vero cells) [9]. The data showed a relatively good correlation with the ricin concentration determined with the ELISA assay. This means that the ricin detected in the samples was also biologically active. Ricin was incontestably identified by means of gel electrophoresis (SDS-PAGE) and western blot.

At first glance, the concentration measured in sample 1 appears to be relatively high. However, in comparison with the concentration in Ricinus communis shreddings that had not been heat inactivated, the sample has a residual concentration of only 3 % of active ricin. This means that 97 % of the ricin is destroyed by heat inactivation. This assessment is based on measurements of ricin concentration in seeds taken from various Ricinus communis cultures [10]. After pelleting, the concentration in sample 2 had been reduced by a further 79 %. This is due to further inactivation through the extrusion process (pelleting) at approx. +80°C. After Ricinus communis shreddings are mixed with rape shreddings to form biological fertiliser and, following the pelleting process, the final result is a ricin concentration in the finished product of 120 mg/kg.

There are no official limits for ricinine content. This insecticide is produced by the plant and its content in shreddings of Ricinus communis is relatively high at 1,700 mg/kg and none is inactivated by heat during the extrusion process. Merely the processing of Ricinus communis shreddings to biological fertiliser has a “diluting” effect that about halves the value.

Conclusion

Measured against the earlier evaluation method set out in the fertiliser ordinance, the ricin concentration is too high. Compared to the corresponding EU ordinance, which was applied until the middle of 2013 in Switzerland too, the value of sample 1 is 16 times too high (< 50 mg/kg). Even in the processed biological fertiliser the ricin concentration is more than twice as high.

At the beginning of November 2013, Oekohum informed the public that, because of the high ricin content, it had been able to return its flawed stocks of Ricinus communis shreddings to the French supplier. A biological fertiliser without Ricinus communis shreddings has been developed as an alternative. Dog owners have withdrawn their demands.

The decision by the Federal Office for Agriculture to prohibit the use of Ricinus communis shreddings for the production of biological fertiliser is a reasonable solution. A regulation with limits always involves the risk that occasionally products with too high a ricin content may appear on the market. If, on the other hand, all producers and importers of fertiliser abide by the prohibition, there should be no more cases in the future of dogs being poisoned in Switzerland by biological fertilisers containing ricin.

It has not yet been resolved what is to be done with the huge amount of more than one million tons per year of Ricinus communis shreddings that ensue as the undesired by-product of castor oil production. This question is related to its possible abuse for terrorist purposes [11]. The ricin contained in shreddings can be extracted with relative ease and processed to form a highly toxic raw product.

Together with saxitoxin, ricin is listed as a Schedule 1 chemical in the Chemical Weapons Convention (CWC) and is thus subject to a comprehensive ban on development, production, stockpiling and use. For this reason, and as OPCW reference lab, SPIEZ LABORATORY studies these two toxins too, along with conventional C agents. Genuine samples like the biological fertiliser containing ricin which was examined on behalf of civilian clients in 2013 are therefore welcome. They provide a practical test and in this way a high level of preparedness can be maintained for examining suspect samples.

Düngemittel: Gefährliche Leckerbissen für Hunde

Bio-Dünger: «Hunde sind wegen seines Geschmacks scharf darauf»

Pflanzendünger ist für Hunde unwiderstehlich. Labrador Harry musste eingeschläfert werden: Er hatte Rizinusschrot gefressen.

Es ist der Sonntag vor Ostern: K-Tipp-Leser Serge Pfändler aus Zuzwil SG ist mit seinem Labrador Harry unterwegs. Auf dem Feld eines kleinen Rebbergs in Schönholzerswilen TG spürt der Hund ausgestreute Pellets auf. Pfändler beobachtet, wie sein Hund davon frisst.

Nur wenige Stunden später erbricht der Labrador – und dies gleich mehrmals. Pfändler fährt am nächsten Tag mit ihm in die Tierklinik Nesslau. Für die behandelnde Tier-

D. SCHÜTZ

Hundehalter Serge Pfändler mit Luna: Labrador Harry (oben) starb an Rizinus-Pellets

Tierhalter Serge Pfändler ist enttäuscht von dieser Antwort: «Schade, dass Bio Suisse hinter Machenschaften steht, die Leben und Umwelt gefährden.»

«Rizinusschrot enthält einen Rest Rizin»

Vertreiberin des Rizinusschrots ist die Ökohum GmbH in Herrenhof TG. Sie hält einen Herstellungsfehler für ausgeschlossen. Geschäftsführer Res Schilling sagt: «Rizinusschrot enthält auch nach dem Erhitzen einen Rest Rizin.» Der Labrador hätte aber eine sehr grosse Menge Pellets fressen müssen, damit eine Vergiftung eintrete.

Pikant: Ganz so harmlos dürfte Rizinusschrot aber doch nicht sein. Bis vor

K-Tipp, May 2013

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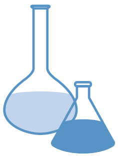


Chemical weapons in the Syrian conflict

Stefan Mogl, Dr. Peter Siegenthaler, Dr. Beat Schmidt

The conflict in Syria and the questions relating to the deployment of chemical weapons dominated the work of the Chemistry Division of SPIEZ LABORATORY in 2013. Spiez was one of the laboratories confirming for the UN/OPCW investigation that the nerve agent sarin had been used. Analytical challenges combined with an exceptionally high media presence characterised the work of the chemists in 2013–2014. For a few weeks, the chemical agent verification lab was in the focus of the public.

What began in March 2011 with demonstrations against an emergency law that had been in force for 48 years, developed into civil war in Syria; and towards the end of 2012 poison gas was allegedly used against rebel positions in the town of Homs. Just half a year beforehand, US president Barack Obama had threatened Syria with military action, if the regime of Bashar al-Assad used chemical weapons or made preparations for their use. "The US government had made it unmistakeably clear to Assad and every player in the region that this would be a red line for us, that it would have huge consequences if we observe movement on the chemical weapons front or their use', the president stated in August 2012.

Were chemical agents used in Syria?
At the beginning of 2013, there was discussion in the international and Swiss media about what substances might have been involved in the alleged attacks of 2012. The Chemistry Division repeatedly answered questions relating to the analysis or the validity of samples, should an on-site investigation become possible.

Generally, in verification analysis – concerning the question of whether chemical agents have been used or not – a distinction is made between the analysis of clinical and environmental samples. The first are urine or blood samples taken from victims (exposed persons) or organ samples from dead animals. Clinical samples should usually be taken within a few days of suspected exposure, as the chemical agent and metabolites (reaction products) formed from it in the body are excreted and can thus either only be detected with difficulty or not at all. When collecting environmental samples, material is taken that has been in direct contact with the chemical agent or, which is most likely, contains a high concentration of agent or its degradation products. Such material includes munition fragments, soil and material samples but also wipe samples taken from

A man holds the body of a dead child among bodies of people activists say were killed by nerve gas in the Ghouta region (august 2013)

surfaces close to the impact point of projectiles where exposure to the agent is suspected. Important for the validity of the samples is an uninterrupted chain of evidence (chain of custody), i.e. the end-to-end documentation of the path of the sample from its sampling point to the analysing laboratory. This ensures that samples cannot be tampered with, undetected, during transport to the analysing laboratory (e.g. by a broken seal).

There was intensive debate in the media about the identity of the substances suspected of having been used. Experts tried to draw conclusions about the kind of substances used from the description of symptoms patients were suffering from in local hospitals. The result of these discussions was very unspecific: Agent 15, irritants as well as nerve agents were considered by observers to have caused the symptoms. The range of substance classes emphasises how difficult diagnosis from afar is, if based solely on a description of the symptoms. Furthermore, one can never completely exclude the possibility that the data transmitted has been manipulated.

In the course of speculation about the identity of the substances that had allegedly been used, the Chemical Weapons Convention (CWC) was occasionally misinterpreted: Certain media claimed that less poisonous substances that might have been used, such as irritants or hallucinogenic compounds, were not chemical weapons. In truth however, according to the CWC every chemical substance employed in a military conflict for its toxic effect is a chemical weapon and its use is therefore prohibited. Such substances explicitly include irritants such as tear gas, even if their use by police forces is permitted for maintaining public order.

Surprisingly, the Syrian government asked the Secretary General (UNSG) on 20 March 2013 to initiate the Secretary General Mechanism. This process allows the UNSG at the request of a UN member state to order an investigation of a possible violation of the chemical or the biological weapons convention. Syria demanded an investigation into the alleged release of chemical warfare agents during a battle on 19 March 2013 in Khan Al Asal. In the following days, France and Great Britain requested the extension of the investigation to include further incidents of alleged use of chemical agents. The UNSG nominated Åke Sellström from Sweden as head of this mission. The Organisation for the Prohibition of Chemical Weapons (OPCW) and the World Health Organisation (WHO) were to accompany this mission at the operational level with their experts. Although mission preparations were rapidly initiated, the team was unable to enter the country, because no agreement could be reached with Syria on access to prohibited sites.

Two months later, the British foreign minister, William Hague, told Parliament in London that Great Britain possessed information that proved that chemical weapons had been used by the Syrian government. And two weeks after that, the French foreign ministry reported that analysis of clinical samples from Syria confirmed that sarin had been used. Later, it became known that the French results were from a blood sample taken after an aircraft attack on Saraqeb on 29 April 2013.

During the same period, the German news magazine DER SPIEGEL contacted SPIEZ LABORATORY. The magazine wished to bring samples from alleged chemical weapon attacks in Syria out of the country through reliable sources and have them analysed by a neutral laboratory renowned for such analyses. SPIEZ LABORATORY agreed to offer its expertise as far as possible and within its competencies, but could not accept the samples because of the absence of a chain of custody. Later, after the sarin attack in Ghouta on 21 August 2013, the magazine criticised in an article that all efforts with various countries, with the OPCW and with the UN to find a laboratory for analysing the samples had remained unsuccessful. While from a scientific perspective it had been right to refuse the samples, the magazine's criticism was not entirely unjustified, because if the UN had not obtained access to Ghouta, these samples could have become important.

On 21 August 2013, DER SPIEGEL contacted SPIEZ LABORATORY again and asked for technical support. An attack had been carried out in the early hours of the morning in the area around Ghouta. Shortly afterwards, videos of people showing signs of poisoning circulated in the Internet. DER SPIEGEL asked for an assessment of the symptoms of adults and children documented in the video material. After analysing the material, SPIEZ LABORATORY gave DER SPIEGEL an interview in the evening of the same day. The article stated that the symptoms of some victims and in particular of children observed in the videos really had been caused by poisoning and were genuine. Furthermore, one concluded that the symptoms indicated poisoning with acetylcholinesterase inhibitors. This can also be caused by nerve agents. This statement by the Chemistry Division led to numerous other media appearances.

After lengthy negotiations with the Syrian government , the UN/OPCW team obtained permission on 18 August 2013 to enter the country and travelled to Damascus. In view of the events of 21 August 2013 and at the insistence of several member states, the UN Secretary General ordered the inspection team to Ghouta, to take samples and collect facts which took place from 26 to 29 August 2013.

On 31 August 2013 the OPCW asked SPIEZ LABORATORY whether it would be willing to analyse the samples of the UN/OPCW mission in Syria. Switzerland affirmed this request.

Analysis of UN samples in Spiez

The samples obtained by the UN mission in Syria arrived initially at the OPCW laboratory in Rijswijk, Netherlands at the beginning of September. There they were prepared for further dispatch to the designated OPCW laboratories. The clinical samples were sent to institutes in Sweden and Finland, while environmental samples were sent to Germany and to Switzerland. Escorted by two OPCW members, the samples for Switzerland arrived on the evening of 4 September 2013 by charter flight in Zurich-Kloten airport. Under arrangements made by the International Relations Division of the Defense Department, the samples were escorted by military security to Spiez.

After receiving the samples, the laboratory first established whether the package and sample container seals were intact, and whether the seal numbers and sample weights agreed with

Textile, soil and liquid samples from Syria

Initial screening of sample containers using handheld C detectors

The analysis of 49 samples (resulting in 230 subsamples) required more than 2400 measurements, using several analytical systems in parallel.

the specifications given in the enclosed OPCW documents. To ensure that neither packaging material nor sample containers were contaminated, chemical agent monitoring was continually carried out with various handheld detectors during the entire unpacking process. After checking the seals and signing the hand-over document, the samples were officially transferred to SPIEZ LABORATORY.

The samples consisted of soil, textile, rubber and hair samples as well as rinsing fluids (extracts) from wipe samples and projectile fragments that the UN inspectors collected in Syria and which had been split up at the OPCW laboratory. In addition, the set of samples contained various specimens of those solvents used in Syria at the OPCW laboratory for taking and processing the samples. Apart from these "solvent blanks" the OPCW provided a soil sample spiked with chemical agent related compounds as a "positive control" to evaluate the analysis laboratory as well as a corresponding not spiked soil sample as "negative control".

As the symptoms of the Syrian victims suggested nerve agent or other acetylcholinesterase inhibitor related poisoning, and the results from unofficial examinations of samples from the Syrian civil war indicated the use of the nerve agent sarin, the UN instructed SPIEZ LABORATORY to analyse the 49 samples primarily for the presence of the sarin nerve agent as well as sarin-related chemicals and to submit a brief summary as soon as possible.

Immediately after accepting the samples – during the night of 4 to 5 September 2013 – the first samples were prepared for instrumental analysis. The goal was to begin analysing the samples on the various analytical instruments of the Analytical Chemistry Branch as quickly as possible. In a first step, the liquid samples were analysed in parallel with nuclear magnetic resonance spectrometry (NMR), gas chromatography mass spectrometry (GC-MS), gas chromatography with nuclear emissions detection (GC-AED) and liquid chromatography mass spectrometry (LC-MS), utilizing several GC-MS systems at the same time.

Only a few hours into the analytical work, sarin was unambiguously detected during the first analyses of samples. After three days of intensive analysis, a first summary report with preliminary results was sent to the head of the UN mission Åke Sellström. It showed that a large proportion of the samples contained sarin and/or sarin-related compounds.

Elaborate processing and analytical work followed using complementary analytical techniques and reference substances to confirm the presence of sarin or sarin-related compounds in positive samples. To definitely exclude the presence of these compounds in the negative samples, further examinations were made with the most sensitive analytical techniques.

About 230 sub-samples resulted from the 49 samples received. Analysing them involved up to nine analytical systems, operating practically round the clock, taking over 2,400 measurements. The greatest challenge of this assignment was the large number of samples and the time pressure which called for efficient time management and expedient setting of priorities. The organisationally complex assignment was also very elaborate in terms of sample and result management and in reporting too – a fact that was reflected in the

The Nobel Peace Prize for the OPCW

Although membership of the Chemical Weapons Convention is voluntary, the OPCW possesses a comprehensive verification regime, which allows monitoring of the compliance with the CWC. At the end of 2013, the OPCW was awarded the Nobel Peace Prize which brought even more world attention to its role. The OPCW received its award “for its extensive efforts to eliminate chemical weapons”. During the 16 years of the OPCW’s existence, some 58,000 tons of chemical agents have already been destroyed under its supervision. This is about 80 percent of declared stocks. The publicity involved with the Nobel Peace Prize should ensure that the OPCW continues to obtain the resources required to fulfil its mandate.

That the OPCW was awarded the Nobel Peace Prize is also a very gratifying milestone in the history of SPIEZ LABORATORY. On the basis of its performance in international analytical OPCW proficiency tests, it was one of the first laboratories in the world to be chosen as early as 1998 as a designated OPCW laboratory.
Today, the OPCW network comprises some 20 designated laboratories that are obliged to prove their competence once a year in OPCW proficiency tests. SPIEZ LABORATORY has been able to achieve a top position in competition with the most renowned institutes of the world. Furthermore, SPIEZ LABORATORY has also significantly contributed to the further extension of the reference library for OPCW on-site analyses (OPCW Central Analytical Database, OCAD).

roughly 1,000 working hours by the six-strong analysis team. The different types of samples as well as the little amout recieved for some samples posed challenges during sample preparation. Furthermore, samples with very high concentrations had to be processed and analysed along with samples containing only traces of sarin-related compounds requiring complex measures for the prevention and elimination of memory effects.

After dispatching the second summary report with refined results on 13 September 2013, the 267-page final report with all analytical data was finished a week later on 20 September 2013 and handed over to the UN mission.

The analyses produced by SPIEZ LABORATORY played a part in enabling the UN investigation commission to present, in its concluding report of 16 September 2013, scientifically irrefutable proof of the use of chemical weapons in the Syrian civil war. The fact that the OPCW/UN entrusted SPIEZ LABORATORY with the Syrian samples, as one of the four laboratories involved, shows that its expertise is acknowledged and that Switzerland’s past support of the OPCW is appreciated.

Syria becomes a state party to the OPCW
On 14 September 2013 Syria submitted its ratification of the CWC to the UN Secretary General and 30 days later – in accordance with the

Chemical Weapons Convention – on 14 October 2013, it became a state party to the OPCW. To prevent Syrian chemical weapons from being used again in the current conflict, the Executive Council of the OPCW took important decisions on 27 September, even before the 30-day period had elapsed. The Council demanded that Syria provide preliminary information on the storage and production facilities for its chemical weapons by the beginning of October 2013 and the destruction of all production or filling facilities by 1 November 2013.

The OPCW immediately began with the inspection of declared Syrian chemical weapons storage and production sites and on 15 November 2013, the Executive Council set out a preliminary schedule for their destruction. Apart from the CW munitions themselves, installations and facilities for CW production – synthesising plants and installations for mixing binary precursor chemicals – as well as storage facilities for chemicals and ammunition were to be destroyed. The practical approach and method of destruction depend on the type of chemical weapon to be destroyed and the Executive Council of the OPCW authorises in advance what method is to be applied and what verification measures are to be taken by the OPCW inspectors (verification).

The OPCW decided to destroy only the precursor isopropyl alcohol on-site in Syria. All other chemical weapons related chemicals – chemical agents, binary substances for their synthesis or precursors – were to be shipped out of the country for destruction because of the on-going armed conflict in Syria. With this approach, the OPCW "broke new ground" – Article I of the CWC prohibits the transfer of chemical weapons and obliges the possessor state to destroy these within the country.

Transporting the chemicals from their storage location to the port of embarkation – a Syrian government responsibility – is/was a critical step due to the on-going civil war and the uncertain security situation. The chemicals delivered to the port of Latakia were checked by the OPCW and loaded onto Danish and Norwegian cargo ships. The head of the Physics Division at SPIEZ LABORATORY, Dr. Mario Burger, senior advisor for the UN Environmental Programme (UNEP), was present on-

Destruction Technicians monitoring flow, pressure and temperature gauges on the Field Deployable Hydrolysis System FDHS

site as technical adviser for environmental issues to monitor the transport and loading process of the joint UN/OPCW mission. The ships then ferried the chemicals to a port in Italy for cross-loading.

Syria declared two CWC Schedule 1 chemicals – one of them is a chemical warfare agent, the other is a precursor. In the Italian port these two substances are transferred to an American ship equipped with a mobile plant developed by the USA for chemical neutralisation (hydrolysis). Afterwards, the chemical mixture, the hydrolysate, obtained through hydrolysis is transferred to an industrial incineration plant.

Via an international call for tenders the OPCW searched for firms that are willing to destroy chemicals or hydrolysate from Syria under the supervision of the OPCW. A Swiss company also figured on the list of 14 applicants published by the OPCW. On 14 February 2014, the OPCW announced that a Finnish company and one from the USA had been contracted for the destruction. The United Kingdom and Germany had confirmed before the tender process that they would destroy parts of the chemical stockpile/hydrolysate at local facilities. The OPCW developed a framework agreement for the verification activities of its inspectors and the Executive Council will decide on what measures are necessary depending on the particular circumstances at each company.

The UN/OPCW mission in Syria has demonstrated the importance of multilateral disarmament organisations. During the Syrian civil war and under the most adverse conditions, independent proof of the use of chemical weapons was provided on the basis of the Secretary General Mechanism. Through Syria's accession to the CWC a further chemical weapons programme will be completely destroyed under the supervision of the OPCW.

The proper management and quality monitoring of individual NBC protection equipment

Dr. Patrick Wick

Proper management and periodic quality monitoring are essential for ensuring that individual NBC protection equipment is in good operational condition. Last year, SPIEZ LABORATORY conducted extensive studies to test the quality and operational performance of the armed forces' individual NBC protection system, of filtering semi-masks and of ABEK-SF04 wide-range filters.

The protection equipment that is exclusively developed by private industry must satisfy high requirements to ensure comprehensive and balanced NBC protection. Before procuring new systems and equipment, tests ensure that the requirements are met. The individual NBC protection equipment of the armed forces and civil protection partner organisations is usually designed for a long service life and stored in such a way that operational readiness is maintained.

The armed forces' Individual C Protection System (ICS 90)

The Individual C Protection System (ICS 90) was fielded by the Swiss Armed Forces at the beginning of the 1990s. It consists of the following main components:

- NBC Respirator 90 (SM 90)
- NBC Filter 90 (SF 90)
- C Protection Suit 90 (CSA 90)
- NBC Gloves 90
- NBC Overboots 90

As commissioned by armasuisse, SPIEZ LABORATORY checked the operational performance of the ICS 90. The condition of the materials used was tested (active carbon, plastics, rubber, textiles) as was the leak-tightness of the Respirator 90 and the overall protective factor of C Protection Suit 90 against gas (VX).

Test results showed that the components of the ICS 90 essentially satisfied the standard required at the time of procurement and can continue to be used. To ensure the quality and operational performance of the ICS 90, SPIEZ LABORATORY recommends periodic testing of stockpiled equipment.

Filtering half-masks (FFP) as a precaution against pandemics

In its current influenza pandemics plan for Switzerland, the Federal Office of Public Health (FOPH) offers the following advice to institutions in regard to filtering semi-masks:

- Ensure that there are sufficient stocks by establishing obligatory stockpiles and minimum reserves.
- Provide health personnel with protective masks through the cantons and employers (hospitals, nursing institutions, home care, etc.).
- Provide employees with protective masks through employers (in companies where employees are exposed to potential risks).

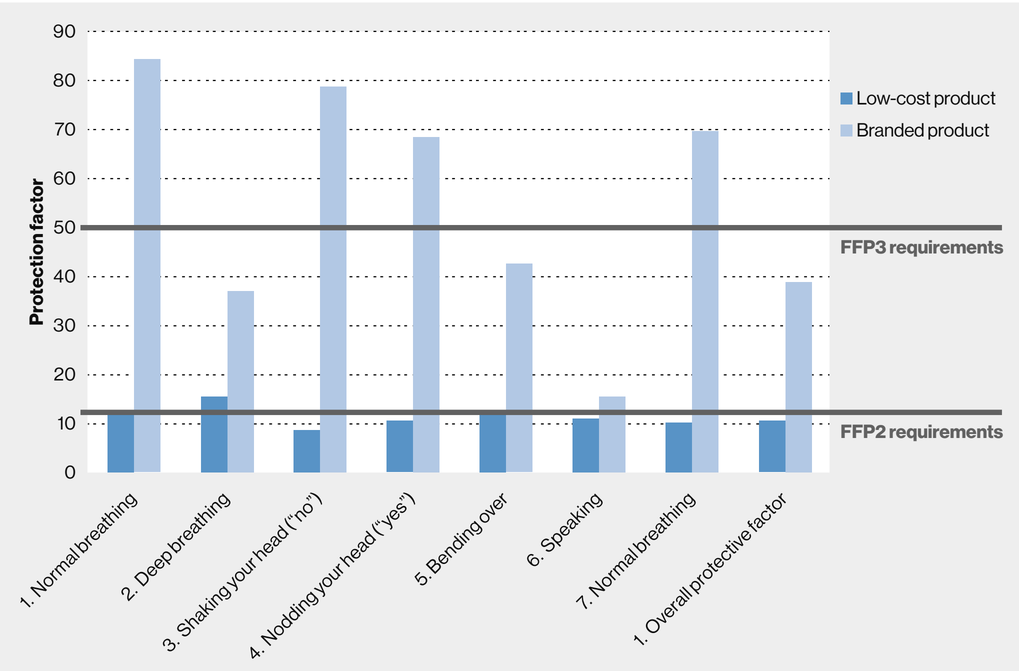
As a personal precaution all members of the population are also required to keep 50 hygienic masks each in reserve.

Corresponding stockpiles were already established during the swine flu wave of 2009. Many manufacturers of filtering half-masks limit the operational performance of their product to 3–5 years. In view of the stockpiles – some of them substantial – held in Switzerland, the question arises as to whether they could be used beyond the expiry date given by the manufacturer. SPIEZ LABORATORY examined two half-masks from different manufacturers to see whether this would be possible.

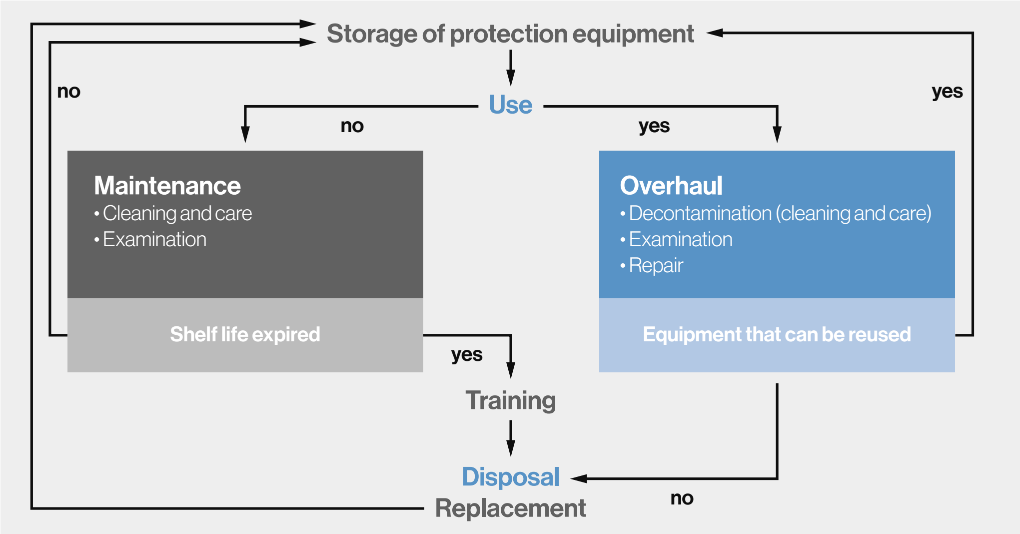
Filtering half-masks: Low-cost product (left) and brand-name product.

A low-cost Asian product and an internationally recognised branded product were examined, both of them filtering half-masks with class 2 exhalation valves (FFP2). In neither case were measurements available from the date of procurement. The examination included checking the condition of the harnesses and how well the masks protected against particles in accordance with the EN 149 standard.

The branded product met the standard requirements without any difficulty, partly even that of the higher class 3. The low-cost product failed to perform as required for FFP2 masks. We recommend replacing the low-cost product with a branded product and making regular visual checks of the condition of the masks and especially of the harnesses.



Leak tightness of the half-masks and requirements according to EN 149. The branded product meets the requirements, partly even that of class 3. The low-cost product failed to perform as required for FFP2 masks.



According to the results, accredited branded products can also be used beyond the expiry date given by the manufacturer.

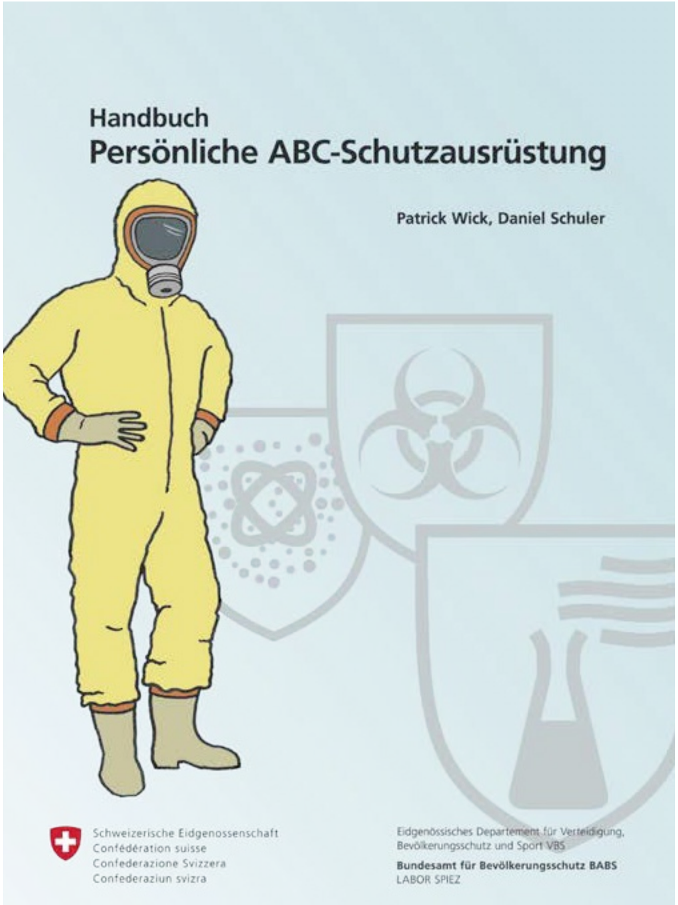
ABEK-SF04 wide-range filter
For civil affairs support operations the Swiss Armed Forces has, in addition to the NBC Filter 90 (SF 90), procured industrial filters (ABEK-SF04) that also provide protection against ammonia and its derivatives. ABEK filters are also stockpiled by civil protection partner organisations. Unlike the SF 90, the ABEK-SF04 wide-range filters are not designed for long-term storage and extended use. Manufacturers therefore limit the operational usability by stating an expiry date (usually after 10 years). SPIEZ LABORATORY tested the operational performance of some ABEK filters that had been stored beyond the expiry date given by the manufacturer: The tested ABEK-SF04 wide-range filters from military stockpiles with expiry dates of September 2010, August 2012 and January 2014 met the relevant absorption requirements. No aging effect could be established. The danger of mechanical damage to the filter casing when carrying it on the body is more critical. Many filters had slight dents.

Despite the good quality of the filters tested, we do not recommend that the armed forces and civil protection partner organisations should build up large stockpiles of them. In normal situations ABEK filter requirements can always be met by industry. With active management, stocks can be kept low and filters in mint condition can always be provided for operations. Despite the pleasing test results, we recommend disposing of filters that have exceeded their expiry date or using them for practice. Training equipment should in any case be designated as such and stored separately from operational equipment.

Proper management of personal NBC protection equipment
Proper management of personal NBC protection equipment comprises of storage and maintenance as well as overhaul after use. Manufacturer's specifications regarding durability, cleaning and care should be taken into account. Apart from cleaning and care, maintenance includes periodic evaluation of stored protection equipment.

Overhaul of protection equipment after operations comprises of decontamination and examination as well as carrying out any repairs. Material that cannot be replaced must be professionally disposed of and replaced if necessary.

Storage influences aging and thus measurably affects the operational performance of personal NBC protection equipment. Constant temperatures and protection against moisture, dust and sunlight (UV radiation) markedly extend the operational life of stored protection equipment. As a matter of principle stockpile turn-over should still be planned so that material in mint condition is always available.



Handbook on personal NBC protection equipment

The range of personal NBC protection equipment (PSA) is huge. Requirements for materials, tests and identification markings have been laid down in standards. Although personal NBC protection equipment is but a sub-area, it comprises about 75 different standards and a large number of widely differing products.

The handbook should provide an overview with understandable and practical information. SPIEZ LABORATORY and its NBC Protection Division have extensive expertise and wish, through this handbook, to address elements with regards to personal NBC protection equipment, whether from the Swiss civil protection organisation or private industry emergency services.

The safe and optimum use of PSA demands both theoretical and practical knowledge, enabling the choice of the right PSA for a certain situation.

The first chapter provides insight into the danger from NBC material: In what form do they arise, what effect do they have on humans, what chemical designations exist and what limits are known.

The second chapter describes NBC protection equipment and is divided into respiratory protection and skin protection.

Basic physical and chemical principles are explained so that the functional mechanism is understood. This understanding makes it possible to deduce what equipment is required for which danger.

Each type of mask, filter or protective clothing is briefly presented, their most important features named and their strengths and weaknesses indicated. Symbols facilitate differentiation between the various items and graphs make it easier to gain an overview.

The third and final chapter provides advice for evaluation, training, management and use of personal NBC protection equipment. On the basis of its operational concepts for defined scenarios a partner organisation is thus able to work out which PSA is most suitable. There will always be a tension between safety and wearing comfort. SPIEZ LABORATORY offers its expertise to the civil protection partner organisations and is available for consultation.

The handbook in German or French can be obtained at: laborspiez@babs.admin.ch

Staff

SPIEZ LABORATORY
Director: Dr. Marc Cadisch[2)]

Secretariat: Irma Lehnherr
Staff: 2 / FTE: 2

PHYSICS DIVISION
Staff: 13 / FTE: 12.8
Head: Dr. Mario Burger[2)]
Markus Astner
Dr. Béatrice Balsiger
François Byrde
Dr. José Corcho
Dr. Emmanuel Egger
Ruth Holzer
Alfred Jakob
André Pignolet
Dr. Stefan Röllin
Hans Sahli
Marc Stauffer
Dr. Christoph Wirz

BIOLOGY DIVISION
Staff: 18 / FTE: 16.4
Head: Prof. Dr. Stephen Leib[2)]
Werner Arnold
Marc-André Avondet
Dr. Christian Beuret
Dr. Olivier Engler
Dr. Rahel Gäumann
Dr. Cédric Invernizzi
Dr. Daniel Kümin
Nathalie Ligeti
Sandra Paniga Rudolf
Jasmine Portmann
Dr. Nadia Schürch
Johanna Signer
Dr. Marc Strasser
Susanne Thomann
Dr. Matthias Wittwer
Fritz Wüthrich
Denise Zysset

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Staff: 15 / FTE: 14.8
Head: Stefan Mogl[2)]
Dr. Beat Aebi
Thomas Clare
Dr. Christophe Curty
Dr. Jean-Claude Dutoit
Dr. Anna-Barbara Gerber
Fausto Guidetti
Roland Kurzo
Dr. Urs Meier
Benjamin Menzi
Dr. Martin Schär
Dr. Beat Schmidt
Andreas Schorer
Dr. Peter Siegenthaler
Andreas Zaugg

NBC PROTECTION DIVISION
Staff: 13 / FTE: 12.3
Head: Peter Hunziker[1) 2)]
Kurt Bachmann
Pia Feuz
Thomas Friedrich
Markus Gurtner
Lukas Gyseler
Marco Hofer
Roland Liebi
Angelo Seitz
Dr. Giuseppe Testa[2)]
Dr. Patrick Wick
Andres Wittwer
André Zahnd

LOGISTICS, QUALITY, SAFETY AND SECURITY DIVISION
Staff: 29 / FTE: 24.6
Head: Mauro Zanni[2)]
Werner Berger
Remo Bigler
Stefan Breitenbaumer
Lisa Brüggemann
Martina Brunner
Werner Bühlmann
Martin Eschler
Béatrice Gurtner Kolly
Daniel Gurtner
Katharina Imobersteg
Felicitas Jegher
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Klaus-Nestor Perrollaz
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René Scherz
Hans Schmid
Isabelle Strasser
Roger Tschirky
Marianne Walther-Leiser
Alexander Werlen[2)]
Marianne Wittwer
Marianne Wüthrich
Rosmarie Zahnd

STRATEGY AND COMMUNICATION
Staff: 1 / FTE: 1
Dr. Andreas Bucher[2)]

DDPS RADIATION PROTECTION TECHNOLOGY
Staff: 1 / FTE: 1
Markus Zürcher

TRAINEES
Staff: 7 / FTE: 7
Miriam Champion
Leonie Gfeller
Bruno Lengacher
Roger Noti
Jan Pridal
Dominik Stettler
Florian Walthert

NOTES
Staff: Total number of employees
FTE: Number of paid full-time-equivalent posts

1) Deputy Director SPIEZ LABORATORY
2) Member of SPIEZ LABORATORY executive management team

Valid 1.1.2014

Organisation

SPIEZ LABORATORY
Dr. Marc Cadisch

DDPS Radiation Protection Authority
Markus Zürcher

Strategy and Communication
Dr. Andreas Bucher

Finances and Controlling
Alexander Werlen

Physics
Dr. Mario Burger

- **Nuclear Arms Control** – Dr. Christoph Wirz
- **Environmental Analysis** – Alfred Jakob
- **Nuclear Issues** – Dr. Emmanuel Egger
- **Radioactivity** – Dr. Béatrice Balsiger

Biology
Prof. Dr. Stephen Leib

- **Biology Arms Control** – Dr. Cédric Invernizzi
- **Bacteriology** – Dr. Nadia Schürch
- **Biosafety** – Dr. Daniel Kümin
- **Research and Clinical Analytics** – Prof. Dr. Stephen Leib
- **Toxinology** – Marc-André Avondet
- **Virology** – Dr. Marc Strasser

Chemistry
Stefan Mogl

- **Chemistry Arms Control** – Dr. Beat Schmidt
- **Analytical Chemistry** – Dr. Peter Siegenthaler
- **Incident Response** – Dr. Beat Aebi
- **Organic Chemistry, Detection, Decontamination** – Dr. Christophe Curty

NBC Protection
Peter Hunziker

- **National NBC Protection and Coordination Office** – Dr. Giuseppe Testa
- **Individual Protection** – Dr. Patrick Wick
- **Collective Protection** – André Zahnd
- **Material Testing** – Thomas Friedrich

Logistics, Quality, Safety and Security
Mauro Zanni

- **Administration** – Felicitas Jegher
- **Engineering Biocontainment Laboratory** – Stefan Breitenbaumer
- **Material Management** – Alexander Werlen
- **Measurement Technology Laboratory IT** – Roger Tschirky
- **Technical Services** – Daniel Gurtner

Accredited activities

ISO/IEC 17025 accredited laboratories	
STS 019	Testing laboratory for the analysis of samples for chemical Warfare agents and related compounds
STS 022	Testing laboratory for adsorbents and respiratory protective filters
STS 028	Testing laboratory for the determination of radionuclide concentrations
STS 036	Testing laboratory for polymers and rubber
STS 054	Testing laboratory for the detection of biological agents
STS 055	Testing laboratory for NBC protection material
STS 101	Testing laboratory for the determination of main and trace elements, their compounds and selected air pollutants

Presentations

Our scientists attend and actively contribute to conferences and offer their input to training courses dealing with NBC protection issues. Below are some of the presentations, given by our specialists during 2013.

Datum	Thema
14.01.2013	Dr. Cédric Invernizzi: Use and Misuse of Biological Agents – The Dual Use Dilemma, EFPL, Lausanne
06.02.2013	Dr. Peter Siegenthaler:The Use of High Resolution Mass Spectrometry and Combined Analytical Techniques for the Identification of Unknown Chemicals at SPIEZ LABORATORY – A Case Study from the 32nd OPCW Proficiency Test OPCW PT-32 Workshop, Den Haag
26.02.2013	Dr. Marc Cadisch: A National Response to CBRN Threats, ESM Meeting, Brüssel
11.03.2013	Stefan Mogl: Die "law enforcement" – Problematik in der CWC: Naturwissenschaftliche Dimension, Auswärtiges Amt DE, Berlin
03.04.2013	Stefan Mogl: The meaning of "produced by synthesis" in the CWC, SAB TWG Convergence in Chemistry and Biology, OPCW, Den Haag
08.04.2013	Stefan Mogl: Report of the Scientific Advisory Board on Developments in Science and Technology for the 3rd Review Conference, OPCW Conference of the States Parties, Den Haag
17.04.2013	Dr. Marc Cadisch: Improving Forensic Capabilities, CSCM World Congress on CBRNe, Dubrovnik
04.07.2013	Prof. Dr. Stephen Leib: Infektionen des Zentralnervensystems, Curriculum Infektiologie Inselspital, Bern
10.07.2013	Dr. Peter Siegenthaler: OPCW Proficiency Testing, DSO National Laboratories, Singapur
13.08.2013	Stefan Mogl: The OPCW SAB, BEC Meeting of Experts, Genf
14.10.2013	Stefan Mogl: Convergence in Chemistry and Biology: Implications for the CWC, University of Bradford, Bradford
22.10.2013	Dr. Martin Schär: Strategische Übungen in Analytischer Chemie, ETHZ, Zürich
18.11.2013	Dr. Beat Schmidt: Kontrolle von Komponenten der Destillations- und Absorptionskolonnen, AG int. implementation meeting, Budapest
04.12.2013	Stefan Mogl: Incapacitating Chemical Agents – Way Forward, OPCW Staatenkonferenz, Den Haag
09.12.2013	Stefan Mogl: OPCW SAB TWG Conference, BWC Staatenkonferenz, Den Haag
26.09.2013	Stefan Mogl: Nicht letale chemische Kampfstoffe, ABC-Schutzkonferenz, Bern
22.10.2013	Dr. Daniel Kümin: How to Choose a Suit for a BSL-4 Laboratory – The Approach Taken at Spiez Laboratory, Jahresvers. ABSA, Orlando
23.10.2013	Prof. Dr. Stephen Leib: ZNS Infektionen – Therapieansätze aus experimentellen Modellen, Universität Basel
31.10.2013	Stefan Mogl: OPCW Open Ended Working Group for RevCon: SAB Report to Developments in Science and Technology, OPCW, Den Haag
01.11.2013	Dr. Nadia Schürch: Biologische Bedrohungen, Blockkurs Katastrophenmedizin, Universität Zürich
15.11.2013	Dr. Matthias Wittwer: Pathogenesis and Evolution of Infectious Diseases, Uniiversität Bern

Publications 2013

The list is not exhaustive. Some of the reports are classified.

General topics, interviews

Sabine Goldhahn
Die dunkle Seite der Chemie
ChemieXtra, 08 April 2013
http://www.chemiextra.com/

Stefan Mogl
Wie beweist man einen Chemiewaffeneinsatz?
Echo der Zeit, 26 April 2013

Stefan Mogl
Möglicher Einsatz von Chemiewaffen in Syrien
Schweizer Radio und Fernsehen SRF, 10vor10, 07 May 2013

Dr. Marc Cadisch
Wir stecken Sie an: CBRN
Informationsschrift KSD 2/13 June 2013

Dr. Andreas Bucher
Unsere Vision ist eine Welt ohne Massenvernichtungswaffen
360°, Das IT-Sicherheitsmagazin von Open Systems, July 2013

Dr. Marc Kenzelmann, Martin Baggenstos
Vorsorge und Bewältigung von ABC-Ereignissen in der Schweiz
CP Crisis Prevention, Fachmagazin für Innere Sicherheit, Bevölkerungsschutz und Katastrophenhilfe, June 2013

Stefan Mogl
Nerve Gas Expert on Syrian Attack: You Can't Fake These Symptoms
Spiegel Online, 22 August 2013

Stefan Mogl
Die Zeit drängt für einen Nachweis von Nervenkampfstoffen in Syrien
Deutschlandradio, 24 August 2013

Dr. Christophe Curty
Alarmantes, ces photos ne donnent que des indices
Le Matin Dimanche, 25 August 2013

Dr. Christophe Curty
Armes chimiques en Syrie: La Suisse propose son expertise
RTS Radio Télévision Suisse, 27 August 2013

Dr. Christophe Curty
Syrie: la guerre empoisonnée
RTS un, 1 September 2013

Stefan Mogl
Die UNO stellte nun zweifelsfrei fest, dass in Syrien Giftgas eingesetzt wurde
Radio SRF 4 News, 17 September 2013

Dr. Marc Cadisch
Es geht um die Ächtung von Chemiewaffen
Basler Zeitung, 12 October 2013

Stefan Mogl
Wir haben über zwei Wochen rund um die Uhr gearbeitet
Der Bund, 2 November 2013

Physics Division

Dr. Emmanuel Egger, Dr. Christoph Wirz
Kurze Übersicht der Kernwaffeneffekte und der möglichen Konsequenzen eines Kernwaffeneinsatzes über einer Grossstadt
Laboratory Note, 20 February 2013

Dr. Emmanuel Egger
Gedanken zur Studie «Mögliche Folgen eines Unfalls im KKW Mühleberg bei ähnlichen Freisetzungen radioaktiver Stoffe wie aus einem Block des KKW Fukushima-Daiichi» des Öko-Institut e.V.
Laboratory Note, 27 February 2013

Dr. Christoph Wirz
Kurzbeurteilung und Erkenntnisse zum Nukleartest Nordkoreas vom 12. Februar 2013
Laboratory Note, 11 April 2013

Dr. Béatrice Balsiger, Dr. Daniel Storch, Stefan Trachsel
KKW-Unfall in der Schweiz: Konsequenzen?
Informationsschrift KSD 1/13 April 2013

Marc Stauffer, André Pignolet
Quecksilber im Boden vor Schützenhäusern auf militärischen Schiessanlagen
Laboratory Report, 18 April 2013

Dr. José Corcho, Dr. Christoph Wirz
Gamma-ray spectrometry as an early and rapid tool in nuclear forensics
Poster Session, 35th ESRADA Symposium, Brüssel, 29 May 2013

Dr. Christoph Wirz, Dr. Emmanuel Egger
Entwicklungen im Bereich nukleare Rüstungskontrolle
Laboratory Note, 5 August 2013

Hans Sahli
Validierung des Hochdruckautoklavs ultraClave IV als Aufschlussgerät zur Bestimmung von Po-210
Laboratory Note, 1 October 2013

Dr. Mario Burger, Alfred Jakob
UNICEF-UNEP technical assistance mission on drinking water quality for the national programme "Villages et Ecoles Assainis", Democratic Republic of the Congo (DRC)
– Report I: analysis of drinking water quality in the Katanga Copperbelt Region – May 2013
– Report II: Analysis of drinking water quality in periurban and rural Kinshasa – June 2013
– Report III: Laboratory capacity assessment for drinking water quality analysis – October 2013

Hans Sahli
Bestimmung von Po-210 in Fischen aus Schweizer Seen
Laboratory Report, 06 December 2013

Biology Division

Pia Müller, Valentin Pflüger, Matthias Wittwer, Dominik Ziegler, Fabrice Chandre, Frédéric Simard
Identification of Cryptic Anopheles Mosquito Species by Molecular Protein Profiling
PLoS One. 2013;8(2):e57486.

Joldoshbek Kasymbekov, Joldoshbek Imanseitov, Marie Ballif, Nadia Schürch, Sandra Paniga, Paola Pilo, Mauro Tonolla, Cinzia Benagli, Kulyash AkyLaboratory Reportekova, Zarima Jumakanova, Esther Schelling, Jakob Zinsstag
Molecular Epidemiology and Antibiotic Susceptibility of Livestock Brucella melitensis Isolates from Naryn Oblast, Kyrgyzstan
in: PLOS Neglected Disease, Feb.2013, Volume 7, Issue 2.

O Engler , J Klingström, E Aliyev, C Niederhauser, S Fontana, M Strasser, J Portmann, J Signer, S Bankoul, F Frey, C Hatz, A Stutz, A Tschaggelar, M Mütsch
Seroprevalence of hantavirus infections in Switzerland in 2009: difficulties in determining prevalence in a country with low endemicity
Euro Surveillance 2013;18(50):pii=20660. Available online: http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=20660

Peltola H, Leib SL
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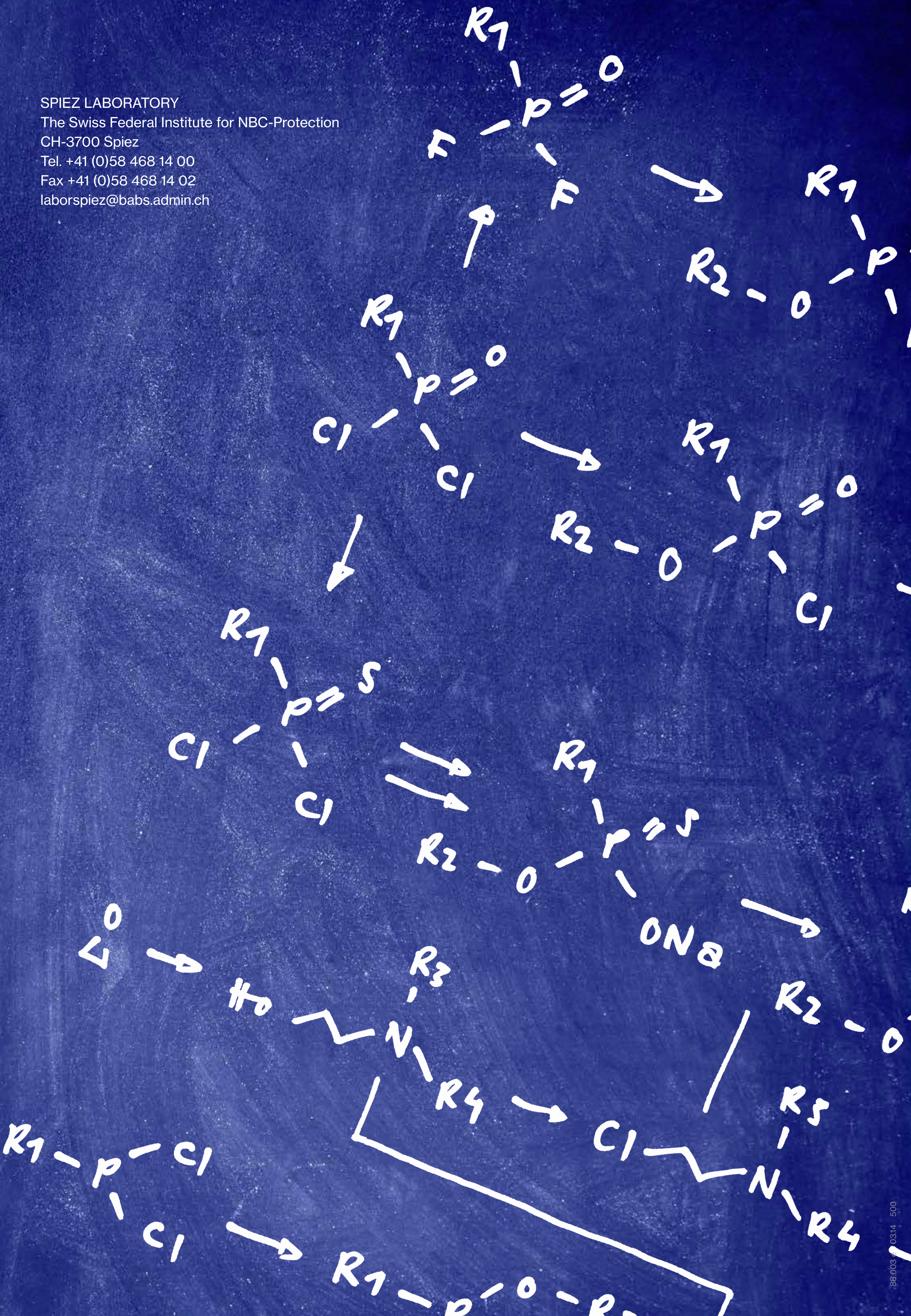
SPIEZ LABORATORY
The Swiss Federal Institute for NBC-Protection
CH-3700 Spiez
Tel. +41 (0)58 468 14 00
Fax +41 (0)58 468 14 02
laborspiez@babs.admin.ch



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