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Biological Experiments on the BION-10 Satellite



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Biological Experiments

on the

BION-10 Satellite

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Foreword

No other epithet could possibly reflect the environment around the Bion-10 project more eloquently than the word *theatre*, a place where the unexpected happens, a place close to, but not necessarily in phase with reality. As long as the reader is willing to accept the total absence of any malignity in this title, we can describe Bion-10 as a play that began in November 1989 and ended in April 1993. This book also deals with the play reviews, the scientific publications, but let's leave those until later.

The Russia Theatre

The theatre - the Soviet-Union, the Russian Federation, the Commonwealth of Independent States, or simply Russia - all of these are name-plates that either decorated the entrance door of our theatre for some time, or were kept in storage to be nailed onto it if needed. Never mind the name changes, more important is that parts of the theatre disappeared with time. One corner of the stage called The Kazakh SSR or Kazakhstan (traditionally reserved for the final act: landing of the satellite) was closed in the process and built up somewhere else under the name of Kazakstan (mind the spelling detail). All of a sudden our play was to change buildings between acts: Bion-10 was to be launched from Russia, but to be retrieved from a foreign country. Another section of the stage was also pulled down. This was The Ukrainian SSR or The Ukraine, where later the Bion-10 scientific debriefing was held: a domestic issue in 1989, but an event in a foreign country in 1993 (called Ukraine, with the same taste for detail).

It goes without saying that such fundamental

changes were accompanied by changes in the theatre management. The flight opportunity offer (the contract proposal to play our role in the Bion-10 play) came to us from a totally different set of people than those who eventually signed up for it. For quite some time we even continued the performance without any stage management at all, and pulled the switches and set the stages ourselves.

The schedule

The scheduling of the different acts was a story in itself. Probably the only forecast that came true was the commitment to confirm the launch opportunity by January 1990 (i.e. to confirm what was announced in a meeting in November 1989). After that, time became something stretchable, apparently inexhaustible, as opposed to something that ticks away and becomes more precious as the hands of the clock progress. Unpredictable and poorly announced delays slipped into the programme from 1990 onwards. Even within the 40-day countdown period, when a number of irreversible biological processes in preparation of the scientific mission were well under way, the launch forecast date was changed three times. The fact that the ESA players were on their feet at Catholic Christmas, Georgian New Year, Orthodox Christmas and Julian New Year reflected the uneasiness of our Russian partners to view time as a progressing line with milestones to be respected, or at least considered.

The stages

Changing the stages was something inevitable in a play that lasted for three and a half years. The main stages in the Russia Theatre were Moscow (our second home), Kújbyshev (which changed its name to Samára), and Plesétsk (the Northern launch site). A few details of each of these stages are worth mentioning.

Moscow, the shop window to the West. The place we had known since 1987 and were on terms with – not that we liked it: we loved it and hated it, in this order and at the same time. Port of entry and port of exit for our personnel and equipment. With no international direct-dial telephones in 1989, but an international operator, the unforgettable 333-41-01, who operated as it would please her (the voice was always female). With rental car contract no.14 taken out with a Soviet-Japanese joint venture. With restaurants permanently closed due to "special services". With desk personnel inevitably absent for lunch at 09:00h in the morning. Even Moscow lacked the minimum infrastructure that was so desperately needed for the job we had to do which is the reason why we built our own house there, Moslab.

Samára, the huge military-industrial complex on the Volga, 1000 km to the East-South-East of Moscow, returned to its pre-revolutionary name early in 1991. A place that was difficult to understand in those days: everything seemed to be secret. Until then inaccessible to visitors from the West, and in this respect the Bion-10 featured a première. In October 1990 the very first two ESA representatives were admitted to what was still called Kújbyshev. Flown from Moscow Vnúkovo airport on a company plane to the industrial airport of Bezymyánka, under the personal responsibility of the host, since no Soviet consulate would issue visas for Kújbyshev. We toured the offices of the Design Bureau "Photon" and the assembly hall of the "Progress" Factory where Soyuz launchers were lined up in large numbers.

Plesétsk, the launch site about 800 km to the North of Moscow, once the busiest kosmodróm of the world and still going very strong in the days of Bion-10. It was also famous because this was the place Francis Gary Powers was heading for in his U-2 spy plane on 1 May 1960, causing an international incident when his reconnaisance aircraft was shot down. This was certainly not going to happen to the Bion-10 cast on its way to Plesétsk on 26 December 1992, but the aerobatic flight and the aborted landing approaches performed by our brave Antónov An-24 in the middle of a blizzard turned quite a few stomachs empty.

Why call the place Plesétsk, if we never even caught a glimpse of the old settlement by the railway track running from Moscow to Archangel that gave its name to the site? In all fairness, it would do more justice to the local hosts if we referred to Mírnyj rather than Plesétsk, since it was in the near-by military town of Mírnyj where we were accommodated. But then, all documentation referred to Plesétsk, whereas Mírnyj – well, there was no map that confirmed its existence.

Leaving aside two short working visits to Plesétsk two months earlier, this trip featured another première: the first payload integration in Plesétsk carried out by ESA and contractor personnel. However honourable it may sound to integrate your own payload, we were taken a little aback when, upon arrival in the integration hall, the Russian operators told us they would rather not touch our hardware. In such cases there is only one thing you can do: take off your shoes, climb into the capsule, bolt up the payload to the right torque value, fix the connectors, make sure to apply loctite, hoist yourself out of the capsule and tell the integration manager your payload is ready for power up! The Russia Theatre, like the Russia House, proved to be an inexhaustible source of suspense.

Wim Jansen

Project Manager for the ESA payload on Bion-10 *De Schelp*, Noordwijk, The Netherlands

1 Introduction

1.1 The Bion Programme

Unmanned, but loaded with assorted animals and plants, recoverable capsules of the Bion type have been used since the early1970s. The goal of these missions was to investigate how the space environment acts on living creatures, with the emphasis on physiological changes in primates and rodents, gravitational biology, radiation biology, radiation dosimetry and radiation protection. To date, more than 20 different biological species have travelled on Bion, including monkeys, rats, bird's eggs, tortoises, tadpoles, fish, insects and insect eggs, caterpillars, plants and plant seeds, mushrooms, cell cultures and microbes. Eleven missions have been completed (Table 1-1).

The scientific management of Bion resides with the Institute for Biomedical Problems (IBMP) in Moscow. As the only consistent biosatellite programme in existence, it has attracted worldwide attention. Already in the pre-Gorbachëv days the doors were opened for participation by non-communist countries. NASA and CNES were actively involved from Bion-3 to Bion-11, while ESA flew payloads on Bion-8, -9 and -10. Over the years, the programme has developed into a true international enterprise with western nations playing increasingly significant roles.

After Bion-11, a smooth continuation of the programme was blocked when NASA decided, under pressure from animal rights campaigners, to stop the research on primates.

In 1999 a new Bion mission – with rats and mice – was considered to focus on radiation biology. The flight was planned to last much longer than previous missions. The spacecraft would be placed in an orbital trajectory similar



to that of the International Space Station (ISS), with the objective of obtaining better information about the radiation health hazards to which the ISS crew is exposed. At the time of writing, this project and other future Bion missions are still under discussion. Figure 1-1. Key IBMP personnel connected with the Bion Project: Prof. Yevgeny Ilyin (right), Project Manager of Bion, and Viktor Golov (left), Chief Systems Engineer.

Table 1-1. The Bion flight log. The term Cosmos is a blanket designation for a wide variety of military and scientific spacecraft. The Cosmos serial number is issued sequentially. By 1 January 2000, 2368 Cosmos payloads had been launched. By way of exception, no Cosmos serial number was attributed to Bion-11, ESA payloads were flown on the marked missions with an asterisk [*]. Bion is also sometimes referred to as BioCosmos, Biosputnik and **Biosatellite.**

Table 1-	1.			
Mission	Also known as	Year of Launch	Date of Launch	Flight Duration
Bion-1	Cosmos 605	1973	31 October	21.5 days
Bion-2	Cosmos 690	1974	22 October	20.5 days
Bion-3	Cosmos 782	1975	25 November	19.5 days
Bion-4	Cosmos 936	1977	3 August	18.5 days
Bion-5	Cosmos 1129	1979	25 September	18.5 days
Bion-6	Cosmos 1514	1983	14 December	4.9 days
Bion-7	Cosmos 1667	1985	10 July	6.9 days
Bion-8*	Cosmos 1887	1987	29 September	12.6 days
Bion-9*	Cosmos 2044	1989	15 September	13.9 days
Bion-10*	Cosmos 2229	1992	29 December	11.6 days
Bion-11	-	1996	24 December	13.5 days

1.2 The Bion-10 Arrangement

The development of joint experiments on the two precursor missions of Bion-10, i.e. Bion-8 (1987) and Bion-9 (1989) was formalised by exchange of letters between ESA and the Interkosmos Council of the Academy of Sciences of the Soviet Union. In both cases the executive body managing the Bion programme was the Institute for Biomedical Problems (IBMP) of the Ministry of Public Health of the Soviet Union. Both precursor missions had been implemented on a no-exchange-of-funds basis, featuring both joint and mutually complementary elements of cooperation from the experiment conception phase up to the publication of the scientific results. In a number of ways, Bion-10 took a different course, mainly due to political changes that led to the demise of the Soviet Union.

Figure 1-2. An overview of the structure of the Bion-10 cooperation.



Whereas the joint payloads on Bion-8 and Bion-9 were still ad hoc undertakings, generated by personal moves and stimuli from the scientific community, Bion-10 was to find its place under an umbrella agreement between ESA and the Soviet Union: Agreement between the European Space Agency and the Government of the Union of Soviet Socialist Republics concerning Cooperation in the Field of the Exploration and Use of Outer Space for Peaceful Purposes. This document, referred to as the Cooperation Agreement and signed on 25 April 1990, mentions inter alia cooperation on joint projects for design, development, launching and operation of payloads, and the conduct of joint experiments in orbit in a number of fields of space activities, such as space biology and medicine, and fundamental research in microgravity. The Cooperation Agreement also reiterates that, as a rule, no exchange of funds is foreseen unless agreed otherwise by the parties, that compensation measures may be worked out, and that projects implemented on a reimbursable basis may be included.

Especially the allowance to include in the implementing arrangement elements of compensation and reimbursement made it possible to extend the cooperation to the Bion satellite designer and payload integrator, the Design Bureau 'Photon' (KB Photon), part of the Central Specialised Design Bureau (TsSKB) based in the city of Kújbyshev (now called Samára). Under the Bion-9 agreement, IBMP had already received compensation in kind in the form of laboratory equipment (and it was to receive much more under the Bion-10 Arrangement), but this time, the industrial counterpart could also be offered a reimbursement that was commensurate with its efforts related to the accommodation of the joint payload.

Following the flight opportunity assignment by IBMP in early 1990, the joint selection of experiments throughout that year, and the signing of the previously mentioned Cooperation Agreement, a draft Bion-10 Arrangement was worked out by ESA and agreed with IBMP and the Interkosmos Council in April 1991. However, in August 1991, in the middle of the approval process on both sides, the *coup* against Gorbachëv heralded the end of the Soviet Union. In December 1991, the Union dissolved itself, and soon afterwards the Academy of Sciences, the Interkosmos Council and the Ministry of Public Health disappeared as well. ESA was left without a partner in the Bion-10 Arrangement.

Fortunately, IBMP as such did not go under in the political turmoil, and the technical and scientific mission preparation continued de facto until other bodies took over the responsibility de jure. In fact, the Government of the Russian Federation replaced the USSR in reaffirming its adherence to the Cooperation Agreement on 28 April 1992, and the recently established Russian Space Agency (RKA) took over the role of the defunct Academy of Sciences of the Union. IBMP was left in its place for the programmatic and technical implementation of the Arrangement. It was later attached to the Ministry of Public Health of the Russian Federation, and later still became a State Scientific Centre in its own right.

The Bion-10 Arrangement between ESA and RKA was signed on 12 October 1992, less than three months before the launch of the satellite. An overview of the structure of this Arrangement is given in Figure 1-2.

1.3 Scientific Cooperation between ESA and IBMP

1.3.1 Selection of experiments

In November 1989, two months after the end of the Bion-9 mission, representatives of ESA and IBMP met in Moscow to exchange ideas about continued cooperation in the field of space biology. Bion-9's successor, Bion-10, had already been announced with a launch scheduled for 1992. ESA presented a list of twelve candidate experiments for this mission.

In a follow-on meeting in Moscow in February 1990, the plans were refined. A shortlist of eight experiments was drawn up. Three were repeats of joint experiments already flown on Bion-9 (DOSICOS, SEEDS and FLIES) and five were new (BONES, MARROW, OBLAST, FIBRO and ALGAE).

Table 1-2. Name **Originator**(s) Affiliation Remarks previously flown on Bion-8 and -9 DOSICOS G. Reitz (D) ESA SEEDS ESA A.R. Kranz (D) previously flown on Bion-8 and -9 FLIES R. Marco, J. Miquel (E) ESA previously flown on Bion-9 BONES J.P. Veldhuijzen (NL) ESA new (Biobox) MARROW G. Schoeters (B) ESA new (Biobox) OBLAST ESA new (Biobox) C. Alexandre (F) M.G. Tairbekov (USSR) IBMP FIBRO new (Biobox) ALGAE H. van den Ende (NL) ESA new CLOUD I.A. Ushakov (USSR) IBMP new WOLFFIA L.V. Nevzgodina (USSR) IBMP new

BONES, MARROW and OBLAST were envisaged to fly together in ESA's Biobox, a programmable incubator planned to make its debut on Bion-10. Two alternative sets of experiments were initially considered for Biobox, namely a cold (22°C) group of experiments and a hot (37°C) group. The hot group - consisting of BONES, MARROW and OBLAST - was eventually favoured because these experiments were not only thermally, but also scientifically, hot. All three investigations were focused on the worrying, ill-understood phenomenon of bone demineralisation in space. The combination of three experiments devoted to this theme was expected to optimise the scientific return from the mission.

IBMP's FIBRO was added as a fourth experiment in Biobox, while ALGAE was to fly as a stand-alone experiment. Half a year later, in September 1990, the shortlist was complemented with two new proposals from IBMP, called CLOUD and WOLFFIA (Table 1.2).

1.3.2 Principal Investigators

The science was envisaged to be 'joint', in the sense that each experiment was to be supervised by a mixed team of investigators whose members were drawn from ESA and IBMP. Joint teams for DOSICOS, SEEDS, FLIES, CLOUD and WOLFFIA were readily formed from scientists who had already worked together on Bion-8 and -9. To support ALGAE, IBMP appointed Olga V. Gavrilova (from Leningrad, later reverting to its old name St. Petersburg), who had previously flown algal cultures for IBMP on Bion-9. Table 1-2. Experiments selected jointly by ESA and IBMP in September 1990 for the Bion-10 mission. Table 1-3. List of Bion-10 investigators. The experiments were supervised by one, two, three or even four Principal Investigators. Whereas FIBRO and OBLAST simply had one, most other experiments had two PIs (one from ESA, the other from IBMP). In some cases the situation grew more complex. FLIES, an experiment that was synthesised from two separate proposals, had two ESAaffiliated PIs. In May 1991, the investigators of SEEDS decided to split their responsibilities in two parts: there was to be one PI in charge of the radiationbiological side of the study, next to another PI taking care of the radiation-physics and dosimetry matters. As a result, FLIES eventually acquired three PIs, and SEEDS no fewer than four. The names of the PIs are printed in bold letters.

Table 1-3.

Experiment

ALGAE

BONES

CLOUD

DOSICOS

ESA Investigators H. van den Ende E. van Spronsen M.L. van den Briel J.P. Veldhuijzen

J. van Loon C. Semeins B. Zandieh-Doulabi

R. Marco

G. Reitz R. Facius R. Beaujean M. Schäfer W. Heinrich

J. Miquel

R. Marco

E. de Juan J. Gonzalez-Jurado M. Maroto

G. Schoeters

C. Alexandre C. Genty S. Palle

I. Bierkens

J. Maes

FIBRO

FLIES

MARROW

OBLAST

SEEDS

011100

WOLFFIA

A.R. Kranz (biology) J.-U. Schott (dosimetry) B. Baican R. Selz

C. Heilmann E. Schopper K. Gartenbach M. Zimmermann

G. Reitz R. Facius

*V.E. Dudkin was later replaced as PI by A.M. Marenny

The recruitment of joint teams for the Biobox experiments took longer. During the second half of 1990 and well into 1991, IBMP searched for scientists to join the teams for BONES, MARROW and OBLAST. A single candidate was eventually found: Natalia V. Rodionova (from Kiev), who was willing to take a share in all three experiments. Eventually, no scientific task could be defined for her in OBLAST, so she became the IBMP investigator for BONES and MARROW, with OBLAST remaining an ESA-only

IBMP Investigators

O.V. Gavrilova

N.V. Rodionova

O.P. Berezovska

I.A. Ushakov

A.M. Alpatov

N. Tarakanova

A.I. Vikhrov

V.E. Dudkin

A.M. Marenny

Yu.V. Potapov

A.B. Akopova

Yu.A. Akatov

V.V. Arkhangelsky

M.G. Tairbekov

A.V. Gabova

L.B. Margolis B.A. Bajbakov

I.A. Ushakov

A.M. Alpatov

N.V. Rodionova

O.P. Berezovska

Yu.V. Potapov N.A. Nefedov

A.M. Marenny

L.V. Nevzgodina Ye.N. Maksimova

V.V. Shevchenko (biology) V.E. Dudkin* (dosimetry)

4

investigation. ESA investigators who were prepared to join IBMP's FIBRO experiment were identified more than once, but the candidates dropped out after a while for various reasons (none of which strictly related to the experiment itself). Like OBLAST, the FIBRO experiment could not be turned into a bilateral undertaking. Table 1-3 gives a list of investigators connected with the joint ESA-IBMP experiments.

1.3.3 Pre-flight meetings

The concept of bilateral scientific cooperation is hard to put in practice when the two parties live typically 2500 km apart, are unable to travel freely to each other's place, speak different languages and are deprived of direct communication via telephone, telex, fax or email. Such was the situation in which the PIs of the joint Bion-10 experiments found themselves.

In the absence of telecommunication, the working meetings gained in importance. In April 1990, such a meeting was held in Madrid with a dual objective: firstly, to exchange the preliminary results from the joint Bion-9 experiments and, secondly, to discuss the plans for the new Bion-10 mission in the presence of the investigators. The meeting was attended by many representatives from the Bion investigator community (Figure 1-3), including a delegation from IBMP consisting of I.A. Ushakov, A.I. Vikhrov, M.G. Tairbekov, V.E. Dudkin and A.M. Alpatov (Figure 1-4).

Another important milestone was the Bion-10 Critical Design Review which took place in April 1991 at ESTEC (ESA's European Space Research and Technology Centre located at Noordwijk, The Netherlands). As in the Madrid meeting a year before, several IBMP investigators met with their counterparts from ESA for in-depth discussions. The mutual tasks of the PIs were further detailed and arrangements were made with the emphasis on sample sharing upon completion of the flight. Whereas the pre-flight preparations were to remain largely one-sided affairs, the post-flight analysis of many experiments could be turned into a true bilateral enterprise. For details on the distribution of tasks within each experiment,



the reader is referred to Chapter 9. Decisions were also taken regarding publication rights. Each PI was free to write and publish a scientific paper independently, without consulting his partner-PI. However, if the paper contained results provided by his colleague, the manuscript needed the latter's approval prior to submittal to a scientific journal.

Four months after the Critical Design Review at ESTEC, representatives from ESA and IBMP met in Leningrad (now St. Petersburg) for the International Biocosmos Symposium (12-15 August 1991) and a meeting of the ESA-USSR Joint Working Group on Space Biology and Medicine (19-20 August 1991). The latter meeting was brusquely disturbed by the coup d' etat against the Gorbachëv administration which occurred on 19 August 1991. The ESA representatives consulted their various consulates and were advised to leave the country as soon as possible. At this moment, it was seriously feared by ESA and IBMP alike that the death knell had rung for the joint Bion programme. If the forces behind the coup had remained in power, this would have most likely been the case.

1.3.4 Post-flight meetings

On completion of the mission, two concluding scientific meetings were held: the Preliminary Data Exchange and the Final Data Exchange.

Figure 1-3. In the absence of telecommunications facilities. working meetings of the investigators assumed major importance. Bion-9 and Bion-10 investigators pose for the camera at the meeting that took place in Madrid, April 1990. From left to right: Greet Schoeters, Paul Veldhuijzen, Christian Genty, Christian Alexandre, Aleksev Alpatov (Head of the IBMP delegation), Ole Rasmussen, Fred Franz (ESA), Peter Segaar, Harry Willemsen (CCM), Dick Mesland (ESA), Roberto Marco, Albert Kranz, Heinz Oser (Head of the ESA Delegation), Tor-Henning Iversen, Ilya Ushakov, Giovanna Lorenzi, Jaime Miquel, Emilio De Juan, (unknown), Miguel Moroto, Günther Reitz.



Figure 1-4. The IBMP delegation that attended the meeting held in Madrid (Spain) in April 1990. From left to right: Viktor Dudkin, Roberto Marco (host), Anatoly Vikhrov, Ilya Ushakov, Aleksey Alpatov, Murad Tairbekov. The Preliminary Exchange took place on 31 August to 1 September 1993 at Kiev, Ukraine, at the Shmalgausen Institute of Zoology, the workplace of N.V. Rodionova (BONES, MARROW). The ESA team consisted of W. Jansen (ESTEC), R. Demets (Hernandez Engineering), J.P. Veldhuijzen (BONES) and J. Bierkens (MARROW). The Final Exchange was included in a later Biocosmos Symposium, which took place 15-18 December 1993. The venue was a river cruise ship, moored at the North River Port of Moscow nearby Rechnój Vokzál. ESA was represented by senior life scientist D. Mesland (ESTEC).

Several investigators from IBMP temporarily joined their ESA-affiliated partners in Western Europe for the post-flight analysis. Within this

Table 1-4. Main parameters ofthe Bion-10 mission.

Table 1-4.

Launch date, time Launch site Separation (entry into microgravity) Orbital inclination Argument of perigee Apogee Perigee Period Re-entry burn (exit from microgravity) Landing date, time Landing site Total flight duration Orbits completed

MT = Moscow Time = GMT + 3h

Sources: KB Photon, IBMP

29 December 1992, 16:30 MT Cosmodrome Plesetsk (62.8°N, 40.1°E) 29 December 1992, 16:38:47 MT 62.8° 105.6° 397 km 226 km 90.4 min 10 January 1993, 06:44:33 MT 10 January 1993 (planned: 12 Jan), 07:16 MT ~100 km north of Karagandá (Kazakhstan) 11 d, 14 h, 46 min 186 framework, I.A. Ushakov (FLIES, CLOUD) worked some months in Madrid, O.V. Gavrilova (ALGAE) in Amsterdam, L.V. Nevzgodina (WOLFFIA) in Cologne, V.E. Dudkin and Yu.V. Potapov (DOSICOS, SEEDS) in Frankfurt-am-Main.

1.4 Mission Summary

Bion-10 was launched from Cosmodrome Plesétsk on 29 December 1992 at 16:30 Moscow time. The spacecraft carried two rhesus monkeys (named Krosh and Ivasha), newts, frogs eggs and tadpoles, fruit flies, desert beetles, silkmoth caterpillars, plants, seeds, seedlings, algae, and mammalian cell and tissue cultures. The main mission parameters are given in Table 1-4.

Bion-10 was an international project with participation from Austria, Canada, the People's Republic of China, Czechoslovakia, ESA, France, Germany, Lithuania, the Netherlands, Russia, Ukraine, USA and Uzbekistan.

ESA was involved in ten experiments, jointly developed with IBMP. Science objectives were to test the influence of weightlessness on cell cultures, insects and unicellular algae, to measure the effects of space radiation on plants and plant seeds and to monitor the space radiation environment.

The total mass of the ESA-provided payload was 56 kg, including the new 42.5 kg Biobox facility. Biobox was readied for flight in Moscow in ESA's Moslab prefabricated laboratory.

Nine days into the mission the temperature inside the spacecraft began to drift beyond its nominal upper limit of 28°C. As the temperature in the capsule continued to rise, eventually reaching more than 31°C, the flight controllers decided to terminate the mission two days earlier than scheduled. After 11.6 days in orbit, the capsule touched down in Kazakhstan, some 800 km east of the planned landing site.

2 The Launch System

2.1 The Soyuz-U Launcher

The Bion-10 satellite was placed into orbit by a Soyuz-U, a sub-type of the stalwart Soyuz launcher which rates as one of the world's most frequently used, most versatile and most reliable launch vehicles. In continuous service since 1966, the Soyuz is qualified for both unmanned and manned missions. In the latter guise, it has been responsible for launching every Soviet (Russian) manned capsule since 1964. ESA astronauts Ulf Merbold, Thomas Reiter and Jean-François Clervoy all flew on Soyuz-U on their way to space station Mir. In July and August 2000, ESA's two pairs of Cluster satellites were placed in orbit by Soyuz launchers. By the end of 2001, the entire family of Soyuz rockets had tallied 1665 launches.

2.1.1 Soyuz-U described

The unmanned Soyuz-U launcher that lifted off from Plesétsk to put Bion-10 into space was 43.8 m long. Empty, the launcher weighs around 30 t while its take-off weight is 305 t (note that the 6.5 t Bion spacecraft is placed in orbit by nearly 300 t of disposable mass). Soyuz is composed of four sideboosters (the first stage), a central booster (the second stage) and an upper portion consisting of the third stage, the payload adapter and the fairing (Figure 2-1). Liquid oxygen and kerosene are used as propellants for all stages.

The first stage

The first stage consists of four boosters that are laterally assembled around the central core. The boosters are cylindrical-conic in shape, with the oxygen tank in the coneshaped portion and the kerosene tank in the cylindrical portion. Each booster is equipped with an NPO Energomash RD-107 engine, with four main chambers and two gimballed vernier thrusters. The verniers provide three-axis control. Like the verniers, the main chambers of the RD-107 are fed by a turbopump powered by gases generated by the catalytic decomposition of hydrogen peroxide in a gas generator. A pyrotechnic switch provides ignition.

The second stage

The core stage is called the second stage, although its engine is fired simultaneously with the first stage. The second stage is powered by an NPO Energomash RD-108



Figure 2-1, Various configurations of the Soyuz-Bion combination from lift-off to touchdown. A. Configuration during lift-off at time L: B. After release of the side boosters at L+120 s; C. After release of the nose cone fairing at L+160 s; D. After release of the core stage at L+310 s; E. In-orbit configuration of the Bion spacecraft beginning L+525 s; F. Bion spacecraft after release of the battery pack prior to re-entry (R-3 h); G. Bion spacecraft after release of the service module (R-30 min); H. Re-entry capsule on landing (R); I. Configuration during lift-off (front view); K. Configuration during lift off (rear view).

Table 2-1.	
$t = 0 \sec \theta$	Lift-off
t = 120 sec	Separation of the four sideboosters (the first stage)
t = 160 sec	Separation nose cone fairing
t = 310 sec	Separation of the central booster (the second stage)
t = 525 sec	Release of the spacecraft; onset of microgravity

Table 2-1. Typical Soyuz launchevents.

engine. This engine differs from those of the sideboosters by the presence of four rather than two vernier thrusters, which are used for three-axis flight control after separation of the first stage.

The third stage

The third stage, which is linked to the second stage by a trellis structure, is powered by a single-turbopump RD-0110 engine manufactured by KB Khimautomatiki. At the turbine exit, the gases are recovered to feed four vernier thrusters that provide three-axis flight control.

Figure 2-2. A Soyuz-U on the launch pad in Plesétsk. (Photo: KB Photon)



The payload adapter and fairing

The spacecraft sits on top of the third stage, its lower part (the service module) sunk into the cylindrical payload adapter, its upper part (the re-entry capsule and the battery pack) covered by the nosecone fairing. Fig. 2-5 shows the Bion spacecraft being lowered into the payload adapter.

2.1.2 Launch events

A typical launch timeline is given in Table 2-1, and the change of configuration during launch in Figure 2-1. The linear accelerations during launch peak at maximally 4.4g (for comparison: 3g on the US Space Shuttle, 14g on a typical sounding rocket). The first stage and the second stage are ignited simultaneously, so that the vehicle is lifted off by the concerted efforts of five boosters. The four sideboosters separate about two minutes after lift-off, leaving the vehicle propelled by the central core for three more minutes. Separation of the second stage is implemented by the direct ignition forces of the third stage engine, which burns for about four minutes until the calculated velocity increment is reached. At this point in time, within nine minutes after lift-off, the spacecraft is released by springs. This event signifies the onset of microgravity. The physical separation of the spacecraft triggers a mechanical command, known as K0 (pronounced as 'kah-nool') by which the scientific payload can be activated immediately, without telecommand, upon reaching microgravity.

Bion-10 was launched on 29 December 1992 at 16:30 local time (Figure 2-4). The launch sequence was recorded by ESA on a cassette tape. The recording shows that five seconds after ignition, the noise of the rocket engines became audible, which indicates that the ESA spectators were positioned at some 2 km distance from the launch pad. The cracking thunder, which gradually faded into a soft rumble, could still be heard for more than three minutes after lift-off. The trajectory of the launcher was such that the vehicle passed straight over the observation post.



Figure 2-3. Two Soyuz-U boosters (first and second stages only) in the MIK integration hall in Plesétsk, 27 December 1992. The launcher is assembled and rolled-out horizontally while lying on a railway wagon shown left. Rotation into the vertical position takes place at the launch pad. (Photo: L. Pieroni)

2.2 The Bion Spacecraft

Throughout the Bion programme the same type of spacecraft was used, although the spacecraft's subsystems have regularly been upgraded. The Bion satellite is derived from the proven Vostok family of manned capsules as used by the Russian cosmonaut Yuri Gagarin. The manufacturer is TsSKB in Samára, with its subsidiary KB Photon responsible for the design. The spacecraft (mass: 6.5 t, length: 6.2 m) consists of three main components: the external battery pack, the re-entry capsule and the service module (Figures 2-5 and 2-6).

2.2.1 Battery pack

The battery pack is a 1.8 m-diameter cylinder, capped by domed ends and attached to the re-entry capsule by four legs. Its silver-zinc batteries provide on average 400 W of electrical power. The cylindrical surface is equipped with hinged shutters which can be opened and closed for trimming the radiated heat. The battery pack is jettisoned before re-entry. Bion-10 carried a large rectangular box on top of the battery pack that contained experimental radiation deflector instruments from Tamara Ya. Ryabova (IBMP).

2.2.2 Re-entry capsule

The re-entry capsule, a 2.2 m-diameter sphere massing around 2.4 t, is the only recoverable part of the satellite. Apart from being recoverable, it is reusable. The capsule used on Bion-10 had flown before in 1989 on Bion-9. The capsule's aluminium alloy structure is covered with resin material for protection against the frictional heat generated during re-entry. This ablative skin is asymmetrically applied, being thicker at the

Figure 2-4. The launch of Bion-10. Bion-10 was launched in darkness on 29 December 1992 at 16:30h local time, but the glow from the rocket motors lit up the entire sky.



Figure 2-5. The illustration shows the Bion-11 spacecraft and its payload adapter. Bion-10 was very similar. (Photo: Yu. Evstratov)

Figure 2-6. In 1987, this model of Bion-6 (Cosmos 1514) was displayed in the space pavillion of "Moscow's Exhibition of National Achievements" expo park. Unlike the real spacecraft, the mock-up was not equipped with KNA external exposure facilities. bottom of the capsule, which points in the ram direction during re-entry. The skin is covered by a reflective layer of aluminium paint, or foil, for shielding against solar irradiation in orbit. This shiny top coat peels off during the re-entry. For optical-thermal reasons, the capsule of Bion-10 sported four horizontal white-painted bands across its circumference (Figure 2-5). Note that, unlike the very similar Foton satellites, the re-entry capsule of Bion-10 was not wrapped in a thermal blanket.

The re-entry capsule (Fig. 2-8) houses the scientific payload, some satellite subsystem hardware, plus the landing parachute which is accommodated in a separate internal compartment. The capsule is equipped with three circular hatches, two on opposite sides for payload installation and removal, with a third hatch giving access to the parachute trunk. On the outer surface of the re-entry capsule of Bion-10, four pan-shaped external containers were fitted (Russian designation: KNA) for exposure experiments (Fig. 2-7). More details on the KNA can be found in Chapter 7.

2.2.3 Service module

The bi-conical service module is 3.2 m long and measures 2.5 m in diameter at the intersection of the two cones. Like the battery pack, the service module is provided with





hinged shutters by which the radiated heat can be controlled. The service module, which is not pressurised, contains the attitude control system, the telemetry and telecommand equipment and the retrorocket. The attitudecontrol system incorporates nitrogen jets and two Earth horizon sensors which it uses to align the spacecraft in preparation for re-entry. The nitrogen is contained in external spherical flasks (Fig. 2-6). While orbiting, the attitude control system is not used, leaving the spacecraft slowly spinning at a rate of about 1 rotation per 10 minutes.

Firing the retrorocket initiates re-entry into the atmosphere by reducing the spacecraft's velocity so that it falls into a lower orbit. After the 30-second retrorocket burn is over, the four retaining straps that keep the service module attached to the re-entry capsule are released. Table 2-2 gives a typical re-entry timeline.



Figure 2-7. A general view of the Bion-10 spacecraft. The 'monkey' experiments were flown jointly by NASA, IBMP and CNES and are not discussed in this report. (Further information about these experiments is to be found in the Final Report of the U.S. Experiments Flown on the Russian Biosatellite Cosmos 2229, NASA Tech. Mem. 110439 (April 1997) Eds. J.P. Connolly, M.G. Skidmore, D.E. Helwig. Moffet Field CA.: NASA Ames Research Center).

Figure 2-8. Cross sections of the Bion-10 re-entry capsule in (a) the X-Z plane and (b) the X-Y plane, showing the location of the experiments flown with ESA involvement. Legend: 1 Biobox; 2 to 6 autonomous experiments including ALGAE, CLOUD, DOSICOS, FLIES, SEEDS, WOLFFIA; BH big hatch; SH small hatch; T4, T5, T6, temperature sensors.





Table 2-2.

R -3h to -0.5h (flexible) Power supply switched from main to auxilliary;	
Battery pack (i.e. the main source) released	d from
spacecraft.	
$R \le -0.5 h$ Auxilliary power supply switched off.	
R -30 min Retro rocket ignited (total burn time: 30 sec):	
Breaking manoeuvre: decelerations < 3.4 g	, .
Service module jettisoned	,,
P 12 10 min Peantry into the atmosphere:	
K-15-10 mm Keening mito the atmosphere,	
Air friction; loss of kinetic energy;	
Decelerations up to $10 g$.	
R -10-9 min Thicker air;	
Free fall, constant speed;	
Low decelerations.	
R -8.5 min Drogue parachute released, 5 sec later:	
Supersonic parachute released:	
Transition from supersonic to subsonic spe	ed
R -8 or - 7.5 min Main parachute deployed:	eu.
Altitude: 2.5 km	
R - 350 msec Landing retro-rocket ignited when spacecraft is	ust a
few metres above ground:	
Deceleration 8.4 g.	
R Impact;	
Nominal shock level 29 g for 40 msec.	

Table 2-3.

components	experiments	mass	envelope
Biobox	BONES FIBRO MARROW OBLAST	42.5 kg	700 × 440 × 378 mm
mini-facilities	CLOUD ALGAE FLIES SEEDS	5.1 kg 3.5 kg 2.4 kg 1.1 kg	270 × 218 × 102 mm Type III Type III Type II
passive packages	WOLFFIA DOSICOS	145 g 145 g	Type I Type I
exposure packages	DOSICOS SEEDS	755 g	see Chapter 7
	Total mass	~56 kg	

2.3 Payload Composition and Accommodation

The ESA-IBMP payload on Bion-10 consisted of Biobox, four mini-facilities (each an autonomous experiment), two small passive packages and a 14-piece set of exposure packages (Table 2-3). The total mass was about 56 kg. The main component was ESA's Biobox, a 42.5-kg multi-user facility that made its debut on Bion-10 (see Chapter 6). The other components, including the mini facilities, were provided by the investigators and their national funding agencies.

Several experiments were encased in standard experiment containers (Table 2-3), referred to as Type I, II and III containers. The dimensions of these containers are given in Table 2-4. The Type I and Type II containers were borrowed from ESA's Biorack project, the Type III containers were European copies of IBMP's BB containers.

Biobox was mounted in the centre of Bion-10's re-entry capsule, between the two monkey seats. The mini-facilities and the two passive packages were placed together on a support plate underneath the seat of monkey Ivasha. The exposure packages were accommodated in the four KNA containers (Figures 2-7 and 2-8).

Table 2-4.

name

Type I

Type II

Type III

 Table 2-2. Re-entry and landing events.

Table 2-3. Composition, mass andsize of the ESA-IBMP payload.

Table 2-4. ESA standardexperiment containers: types,dimensions and volume.

Biorack Type I/0 Biorack Type II/0 BB

also known as

envelope (incl. appendages)

90 × 58 × 24 mm 100 × 70 × 67 mm 176 × 157 × 114 mm

internal volume

68 cm³ 345 cm³ 2274 cm³

3 Operations and Logistics

3.1 Mission Operations Sites in Moscow

3.1.1 IBMP Khoroshëvskoye

The mail address and telex and fax numbers of IBMP refer to what we would call the 'Head Office' (IBMP staff would rather call it by the name of the nearest underground station: Khoroshëvskoye). There is a control room on the site with communications to the Flight Control Centre (TsUP) in the south of Moscow, and where, during a Bion flight, television images of the orbiting monkeys can be received. For Bion-10, the Head Office was only occasionally used for meetings.

3.1.2 **IBMP** Shchúkinskaya (the School) The second location in Moscow, which originally belonged to IBMP, was its Radiation Research Department, housed in a former school building in the Shchúkinskaya Street, hence known as 'the School'. At the School, the exposure experiments for Bion-10 were mounted on their support plates. Personnel from the School were also responsible for bringing the plates to Plesétsk, and for the installation of these plates in the KNA exposure facilities. After flight, the exposure experiments were subjected to an inspection at the School before being returned to the investigators.

In October 1990, part of the School split off from IBMP to become a research institute in its own right, still under the wing of the Soviet Ministry of Public Health. This new institute was called the 'Research Centre for Spacecraft Radiation Safety', abbreviated to RCSRS. The other part of the School remained affiliated to IBMP, but now under the new designation 'Radiation Safety Service for Spaceflight', abbreviated to RSSS. As Siamese twins, the two similarly-named institutes continued to use the School building together as their place of work, and investigators from both RCSRS and RSSS were jointly involved in the experiments SEEDS and DOSICOS. The institutes later found separate scientific niches, RCSRS specialising in ground-based dosimetry in connection with ecology and radiation pollution and RSSS pursuing radiation dosimetry in space.

3.1.3 IBMP Plánernaya

Out of the three IBMP sites in Moscow, one was extensively used by ESA for pre- and post-flight work and for activities during the orbital flight of Bion-10. This was Plánernaya, named after an old airfield for gliders, situated along the motorway from the international airport of Sheremétyevo-2 towards Moscow, just before where it crosses the motorway to St. Petersburg (Figure 3-1). This establishment was built in the early 1970s. It has no apparent mail address or street number.

Figure 3-1. The IBMP Plánernaya establishment.



Access to the site is restricted to visitors announced with sufficient notice.

The relatively vast surface area of Plánernaya is occupied by buildings identified by number, among which 'Building 4' gained a reputation as the laboratory for Bion-9 in 1989. During the Bion-10 mission Building 4 was the site where the fruit fly experiments FLIES and CLOUD were prepared, and where the 1g ground control experiments for the non-Biobox experiments (ALGAE, CLOUD, DOSICOS, FLIES, SEEDS and WOLFFIA) underwent, in a programmable incubator, a near-synchronous replication of the temperature history of the flight experiments. Next to Building 4 stands a towering complex, once designed for work related to the Buran orbiter [1]. Plánernaya also features laboratory space for primate and rodent research, offices, a nicely equipped space museum and a canteen (which, during the days of economic hardship during the Bion-10 campaign, was run as a bakery). Located between Building 4 and the side gate of the compound, Moslab was erected in May 1992 (see Section 3.2).

3.1.4 TsUP: The flight control centre The flight of Bion-10 was controlled from a military flight control centre in Moscow, referred to as TsUP [2] and located next to the well-known IKI institute. The TsUP received its information through a network of ground stations. A subset of the spacecraft telemetry, related to Biobox, was relayed from TsUP to ESA's Moslab (see Chapter 8).

3.1.5 Moscow State University

The component units of the FIBRO experiment were loaded at the Moscow State University and, from there, transported to Moslab for integration with Biobox.

3.1.6 Novotel

The ESA personnel, the contractors and most of the investigators lodged in the Novotel at Sheremétyevo-2 airport, one of several brandnew, western-style hotel hotels that suddenly appeared in Moscow after the Soviet Union was dismantled.

3.2 Moslab

3.2.1 The history of Moslab (1992-1995) The design, construction and installation of ESA's Moscow Laboratory, known as 'Moslab' (Figures 3-2 and 3-3) represented ESA's first, and to date only, building activity in Russia. Moslab was ESA's first outpost in Russia [3].

In January 1990 ESA obtained from IBMP of the then Soviet Ministry of Public Health the opportunity to fly joint experiments on the Bion-10 satellite, to be launched by the end of 1992. As work progressed through 1990, it soon became clear that dedicated laboratory and engineering facilities would be needed for pre-launch and post-landing work on the payload. This applied in particular to the complex operations with Biobox, ESA's first multi-user facility for microgravity experiments to fly on a Russian spacecraft. The Bion-10 programme was carried out on a no-exchange-of-funds basis between ESA and IBMP, with compensation in kind for the engineering and operational services provided to ESA by IBMP and its industrial partners. Moslab and its associated laboratory and office equipment became a logical part of the compensation package that was agreed upon by both parties (see also Figure 1-2).

Figure 3-2. ESA's Moslab building.



The first exploratory discussions on the feasibility of Moslab were held in the spring of 1991 between the ESA project team and engineers of the Dutch company Wagenbouw (now called Wb Accommodatie Service). The contract was signed in July 1991, and on 27 May 1992, ESA took delivery of Moslab as a fully operational laboratory in Moscow, on the premises of the Plánernaya establishment of IBMP. Moslab was fully equipped for life science and other payload preparations, with its own kitchen and office facilities, and (at that time) unique direct-dial telephone and fax links via satellite.

During the 36 months between May 1992 and May 1995, Moslab was ESA's home base for four spaceflight campaigns: the Biopan-0 test flight on Foton-8 in October 1992, the Biobox-1 flight on Bion-10 during December 1992-January 1993, the Biopan-1 flight on Foton-9 during June and July of 1994, and the Biobox-2 flight on Foton-10 during February and March of 1995. During this time, ESA maintained its ownership of Moslab, but used it only for the specific purpose of mission work on unmanned Russian spacecraft. Between missions, the building and its facilities were used by IBMP personnel.

Designing a laboratory in The Netherlands for installation as a fully operational unit in Russia was not an easy job. Only those who were directly involved in the creation of Moslab remember the difficulties encountered at all stages of the design, construction, export, transport and installation. Between the beginning of the design activities and the departure of the transport convoy to Moscow, the Soviet Union disappeared, and with it the Union's once ambitious space programme. Uncertainties as to the longer-term future of Moslab sneeked into the programme. With time, the situation deteriorated, and maintaining Moslab by remote-control became an untenable financial burden rather than an asset. After the Foton-10 mission it was decided to hand over the property rights of Moslab to IBMP.

3.2.2 Description of the Moslab facilities Essentially, Moslab was designed as a



prefabricated laboratory unit comprising six modules, with the different working areas on either side of the central corridor. The total useful area was 108 m².

The first room on the right-hand side after entering the building was the engineering room. This room was primarily designed for all inspections, tests, checkout, integration and de-integration of flight and ground control hardware during a mission campaign. Tables along two walls provided the countertop space necessary, with shelves and cabinets available for stowage of small hardware, documentation and stationary. Additional countertop and stowage space was provided by two rollaround tables with drawers.

Next in the row was the central laboratory. This area, which included a sink and two work benches, was designed and equipped as a general-purpose laboratory. A big laboratory refrigerator-freezer combination and an autoclave were also located here.

Further down, still on the right-hand side, was the entrance to the microscope laboratory, a small working area featuring a vibration-free microscope table mounted on a steel platform that rested on the concrete foundation of the crawling space under Moslab. Through the microscope lab one entered the cleanroom. This was not a cleanroom in the strict laboratory sense of the word. Rather than pretending a particular cleanliness standard for Figure 3-3. Moslab.

the room itself, the name referred to the two laminar cross-flow benches housed in the room, along one side-wall. The cleanroom was to be used for handling equipment in a reduced dust atmosphere. In addition, biologists could use the sterile atmosphere of the benches by switching on the incorporated UV lights before work.

Down the far end of the corridor was the vivarium, designed to house laboratory animals or plants, with a rack for mice or other small animals, and wall-mounted shelves to hold any accessories that would be required. The lights in the vivarium were controlled by a timer to give the appropriate light/dark regime.

In the same area, but on the left-hand side was located the radioisotope laboratory, later called the hazardous chemicals laboratory. This area was equipped with two laminar down-flow benches, certified for hazardous operations and, in particular, for working with radioactive substances. The rate and direction of the airflow in such benches are chosen to reduce the radiation and contamination hazard to a minimum. Also in the room was an explosionproof refrigerator for storing radioactive or hazardous substances. Access to the radioisotope laboratory was through a lockable airlock with a separate water supply and sink.

Coming back along the same side of the corridor, and passing the toilet, one came to the office-kitchen, which was used primarily for meetings, seating eight-twelve around the tables. One part of the room served as a communications centre and contained the telephones and telefax equipment, including the satellite links abroad through one of the Moscow overlay networks. An overhead projector, a white board and plenty of stationary were available for meetings. The office-kitchen was the only area of Moslab where smoking was permitted and the only place where food and drink were allowed. These items were stored in cupboards and in the kitchen refrigerator, and could be made ready (cooking utensils, ceramic hotplate and coffee machine available) and consumed

(dishes, cups and utensils) in this area. Cleanliness standards and habits valid in ESA member states prohibited the presence of any smoking, eating or drinking material anywhere else in the laboratory and imposed certain minimum rules of hygiene to be adhered to by all users throughout the building.

To complete our tour of Moslab, but without going into details, we should mention the cloakroom, the utilities room (both located near the entrance) and the crawling space, which stretched out under the entire floor surface of Moslab and which kept the working quarters dry during the uncomfortable rise of the water table during spring time. To cope with the harsh Russian winters, the insulation and central heating were designed for a temperature difference between inside and outside of 60°C. To withstand the hot continental summers, a slightly sloped and reflective sun roof was permanently installed over the flat top roof, providing a constant flow of ventilating air between the flat top and the sun roof. This had the additional advantage of keeping the snow off the flat-topped main roof during the winter. Wheeled airconditioners could be installed in all areas with windows, to provide additional cooling during hot periods of the year.

3.2.3 How Moslab performed

Without Moslab, the Bion-10 Mission could not have been accomplished. This statement is so short and uncompromising that it deserves no further elaboration but it still poses the question of how did the facility perform in practice?

The working space turned out to be sufficient but not over-generous. During the peak period of experiment preparation, just prior to transport to Plesétsk, some areas in the building were too crowded for work that required a high level of concentration. The experience gained from this mission, together with that of its immediate predecessor (Foton-8 with the Biopan-0 payload), made it clear that a combined Biobox-Biopan payload could not be handled in Moslab, unless extensions to the facilities were made. The equipment in Moslab fulfilled its purpose without suffering any major breakdowns.

The kitchen and office area was marginal in size, but functional and indispensable under the circumstances, since IBMP had no canteen available and there were no other catering services in the area other than those at the international airport or in the hotel. Shopping, cooking and dish washing were daily chores for the participants.

The central heating system of Moslab worked perfectly and all inhabitants were grateful that it was overdesigned. On New Year's eve 1992-1993, just before the stand-by crew left the premises, the external thermometer reached -30°C, the limit of its measurement range where it got stuck. The moderately heated cellar of Moslab maintained a comfortable constant temperature, an ideal environment for calibrating the temperaturecontrolled transport containers and their travelling temperature loggers!

3.2.4 Moslab: concluding remarks

Moslab remained ESA's Russian bridgehead in its partnership with IBMP for 36 months. During this time, four major operations conducted from Moslab forged a special bond of friendship and solidarity among the engineers and scientists involved, isolated as they were from the rest of ESA and exposed to the uneasy post-Soviet environment. However, as a social experiment, Moslab came too early to become a lasting asset for ESA. Starting with Foton-11 (1997), all payloads for unmanned missions on Foton satellites were to be prepared and integrated at ESTEC and then flown from The Netherlands to the Plesétsk launch site.

3.3 Plesétsk

3.3.1 Kosmodróm Plesétsk

The existence of the launch site at Plesétsk was not acknowledged by the USSR until 1983, although Western observers had long noted that some Soviet spacecraft, judged by their orbital parameters, must have been launched from a site in northern Russia. The Kosmodróm at Plesétsk owes its name to a nearby small-town railway station on the railway from Moscow to Archangel.

Western visitors were, and still are, only sporadically granted access to Plesétsk. ESA personnel were not admitted to the Kosmodróm until 1992, although the Agency had scientific payloads launched from Plesétsk from 1987 onwards. The secretive atmosphere that one sensed in Plesétsk added a genuine flavour of adventure to the mission campaign.

3.3.2 Mírnyj

Tucked away in the endless forests of northern Russia, close to the banks of the river Yemtsá, lies the military base of Mírnyj, a perfectly rectangular pattern of perfectly rectangular buildings in true Soviet style. The base owes its very existence to space and you may still hear the tales of pioneering and hardship that go back to the late 1950s. A nicely equipped and decorated museum on the central square tells you all about it.

The airfield appeared to be a quiet military facility, with just a few Antonov An-2 utility biplanes and an occasional Antonov An-12 cargo plane parked here and there. The Zaryá hotel, on Lenin Street, provided more than adequate accommodation, in huge and well heated apartments for one or two occupants each. There seemed to be no shortage of food either in the restaurants or in the shops. The mission participants and their equipment were shuttled back and forth by bus. The sparse traffic moved slowly on Mírnyj's snowcovered streets and the little noise it made was attenuated by the omnipresent snow.

3.3.3 The MIK

An hour's drive along bumpy roads from Mírnyj was the Assembly and Test Complex (MIK) that had been assigned to the Bion satellite and its payload. The MIK is easily described as an enormous hall with a few railway tracks running from one end to the other; integration platforms are arranged along one long side of the building and offices along the other. Trains loaded with rocket stages and satellites would enter through one gate in the



Figure 3-4. A sketch of the area around the launch site at Plesétsk.

short side and, after completion of payload integration, another train would roll the fully assembled launcher-payload system through the opposite gate onto the selected launch pad a few miles away. The working atmosphere in the MIK was surprisingly quiet and almost relaxed, no doubt the result of years of training and experience on standardised hardware with an impressive success record that gave nobody any reason for concern.

3.3.4 The launch pad

The launch of Bion-10 in total darkness (Figure 2-4) and bitter cold was witnessed from a point along a country lane between the pad (Figure 3-5) and the MIK, where hot tea and rolls were kindly provided by the military to the spectators in an open wooden booth.

Plesétsk or Mírnyj was a unique experience. To be able to tell people at home where we had been and what it all looked like, we went to a great deal of effort to identify all possible landmarks to compose our first map of the site which, with all its inaccuracies, was still infinitely more detailed than anything that could be found in the press. It is reprinted here as a souvenir (Figure 3-4). Plesétsk will always be remembered as an oasis of tranquillity and hospitality.

3.4 **Pre-Flight Operations**

3.4.1 Experiment preparations in western Europe

A minority of the experiments was prepared in Western Europe: the SEEDS packages were integrated in Germany at the J.W. Goethe University, Frankfurt-am-Main, and from there carried to Moslab. DOSICOS (the ESA part of it) was also assembled in Germany, at the DLR in Cologne. The other experiments were, wholly or partly, prepared in Moscow, with investigators from all over Western Europe gathering in Moslab (Figure 3-6).

3.4.2 Experiment preparations in Moscow

The main experiment integration site was ESA's Moslab, where the two Biobox models (flight and ground) were readied for the mission. The Biobox experiments BONES and MARROW were entirely prepared in Moslab. By contrast, the experiment units of OBLAST and FIBRO were pre-loaded at the investigator's home institutes (LBTO/St. Etienne and the Moscow State University respectively) and then delivered to Moslab for integration with Biobox. Moslab was also the place where the ALGAE experiment underwent its final integration and check-out. Next door, in IBMP's Building 4, the fruit fly experiments FLIES and CLOUD were made ready.

The experiments WOLFFIA and DOSICOS (the Russian part of it) were prepared elsewhere in Moscow, at the School. The integration procedure of the exposure experiments at the School is detailed in Chapter 7.



3.4.3 Logistics in Moscow

Rental cars were used to shuttle between the hotel and Moslab, a 10-minute drive. The cars were also used to reach remote sites in Moscow such as IBMP Headquarters, the School and the University. Communication between Moslab, the hotel and the other sites was by telephone. Both Moslab and the Novotel were equipped with direct international telephone lines.

The satellite phone and fax facilities of Moslab were invaluable throughout the entire mission. What a change with respect to the Figure 3-5. The Soyuz-U launcher that carried Bion-10 into space, standing on the launch pad at Plesétsk at about 15:15 (L-75 min), 29 December 1992.



Figure 3-6. The geographical locations of the investigators' home institutes, plus Moslab and Plesétsk.

Bion-9 campaign in 1989, when we relied on the goodwill of a trading company housed in the same hotel as the ESA staff for providing us access to their precious fax, or the Foton-7 campaign in 1991 when we constantly shuttled between the lab and the business centre of another hotel for international communications! How painful it was to revert to 'hotel links' during the days we had a total communication black-out (6-8 January 1993), which, in strict compliance with Murphy's Law, were the days the spacecraft ran into its temperature crisis.

The feasibility of using mobile (cellular) phones, a novelty in 1992, was explored by ESA, but rejected when the pioneering provider pointed out that transmissions from and to Moslab were impossible owing to signal distortion from an electricity generating station in the vicinity of Moslab.

3.4.4 Lead time to flight

IBMP had undertaken to inform ESA of the launch date 40 days in advance. However, things did not proceed as simply and straightforwardly as had been hoped. Within the 40-day period preceding the launch, ESA received notices of three shifts in launch date. The final, and actual, launch date was announced just 20 days in advance:

- on 6 November 1992 the first notification arrived at ESTEC, stating that Bion-10 would be launched between 20 and 25 December, with the exact date to be confirmed before 24 November;
- on 23 November, IBMP declared that the most probable date for the launch of Bion-10 was 26 December (advance notice: 33 days);
- two weeks later, on 7 December, IBMP announced that the launch had slipped to 30 December (advance notice: 23 days).

 two days later, on 9 December, ESA was notified that the launch date had been shifted back to 29 December (advance notice: 20 days).

The frequent re-scheduling caused a multitude of problems in the areas of travel arrangements, visa application and experiment preparation. In fact, by the time ESA received the fourth notification, BONES had already passed its "point-of-no-return" from where amendments to the timeline were no longer allowed (to obtain foetal bones of the correct developmental stage at launch, the mother mice had to be fertilised earlier than 20 days before launch).

3.4.5 Plane or train

The experiment preparations in Moslab started two weeks before launch, and were planned to be closed out on the morning of Saturday, 26 December, whereafter the payload, the ground support equipment and the accompanying personnel would be sent off to Plesétsk. Integration with the spacecraft was planned for the evening of 27 December, two



days before launch. A special aircraft would be made available for the 800-km trip from Moscow to Plesétsk, as no scheduled flights were available for this route. A cause of considerable concern to ESA, which lasted well into the countdown phase, was that IBMP could not confirm that this aircraft would actually be available. Figure 3-7. Trolleys and thermal boxes in the MIK. The autonomous experiments were transported from Moscow to Plesétsk in thermal boxes (shown right). During transportation the boxes were carried on trolleys (shown left) and were powered by batteries contained inside the blue packing cases.



Figure 3-8. The Bion-10 spacecraft in the MIK on 27 December, shortly before integration of the payload.



Figure 3-9. Integration of the Bion-10 payload in the capsule, 27 December 1992. The shiny silvery case is Biobox. The three blue-grey boxes on the floor panel contained the FLIES (3F) experiment, ALGAE (1F) and SEEDS-DOSICOS-WOLFFIA. The white box on the floor panel is CLOUD. The insulating foam padding (right) covers the parachute compartment. On 18 December, with the date of departure to Plesétsk uncomfortably near but still without guaranteed transport by air, ESA was requested by IBMP to prepare for a fallback option: to take the train from Moscow to Plesétsk, which was on the line to Archangel. An ESA/IBMP team went to the railway station to check the timetables and to verify that the voluminous cargo would fit into the train. It would not. This was demonstrated by an accommodation check, in which ESA vainly attempted to manoeuvre a bulky Biobox transport container through the wagon's narrow gangways. After consultation with the railway personnel, it was understood that the cargo could be taken to Plesétsk by train, but only if it were transported in a specially assigned freight wagon; however, surveillance of the cargo during the 16-hour train ride was not possible. Although highly unattractive, the railway alternative was ultimately deemed feasible. Armed with train tickets purchased at the Intourist office in the Novotel, ESA was prepared for a possible emergency in case the aircraft did not show up.

3.4.6 Into the blizzard

Eventually, the availability of the aircraft (an Antonov An-24 twin-propjet) was confirmed. But this did not mean that the worries were over. On the very day of departure, 26 December, the airfield of Plesétsk was closed due to a violent snowstorm and consequently, the Antonov was not permitted to leave Moscow. After several hours of waiting on Vnúkovo Airport it nevertheless took off but on approaching Plesétsk in darkness, it flew into the blizzard. The An-24 trembled and shook ferociously. A lamp broke loose from the cabin ceiling. Many seats in the aircraft were not equipped with safety belts, making the outcome of the flight very unpredictable. This adventure was worse than a ride on a roller-coaster. Conditions were so difficult that it took the pilot three approaches before the aircraft was properly positioned and sufficiently stabilised to touch down on the ice-covered runway of Pero airfield, Plesétsk. That the flight took place and ended safely is a credit to the pilot's skill.

3.4.7 In the MIK

The night was spent in hotel Zaryá in Mírnyj, with the flight hardware and the support equipment temporarily parked underneath the stairway. The IBMP investigators from FLIES and CLOUD split off to the 'Hut', a wooden building at the outskirts of Mírnyj (Figure 9-5). In the Hut, the CLOUD device underwent its final check-out and programming. The next day, 27 December, the ESA cargo and personnel were transferred by bus to the Assembly and Test Complex (MIK).



As mentioned in the Foreword, contrary to the earlier agreed plans, the integration of Biobox with the spacecraft was not carried out by the Russian technicians. Upon entering into the MIK, the ESA personnel and contractors were kindly requested to do the job by themselves (Figure 6-9). By midnight, the integration of the payload was finished. For an overview of the operations timeline during the three days preceding the launch, see Figure 3-10.

3.5 Operations During Flight

3.5.1 Biobox

During flight, the behaviour of Biobox was continually followed in Moslab via a modem link with the TsUP, as more fully described in Chapter 8. Meanwhile, 1g reference experiments were conducted in near-synchrony in Moslab, in a duplicate model of Biobox. More details are given in Section 6.6.6.

3.5.2 Autonomous experiments

The ground reference experiments of ALGAE, CLOUD, DOSICOS, FLIES, SEEDS and WOLFFIA were conducted in an ESAprovided programmable thermal chamber. In this chamber, the fluctuating onboard temperature, as followed on Earth by telemetry, was replicated. As no real-time information about this temperature was available, the ground control experiments were not conducted in synchrony, but delayed 24 hours with respect to the flight experiments. The chamber was operated by IBMP personnel and was located in IBMP's Building 4, next to Moslab. Figure 3-10. The Bion-10 preflight operations time schedule. The close-out of the payload preparations in Moslab took place more than three days before launch. One single experiment, CLOUD, underwent its final preparations at Plesétsk, two and a half days before launch. A CLOUD-like scenario was considered for Biobox, but was rejected because of the poor infrastructure and lack of laboratory facilities at Plesétsk. Note the one-day time span between the arrival of the payload at Plesétsk and the start of integration of the payload into the spacecraft. This delay was to be significantly reduced for Biobox-2 of Foton-10, when permission was obtained to travel to Plesétsk on the very day of integration. Integration of the payload with the spacecraft was completed less than two days before launch. The rotation of the satellite, from vertical to horizontal, marks the close-out of satellite preparations and the start of the mating the satellite with the launcher. Less than one day elapsed between the installation of the launcher on the pad and lift-off.



Figure 3-11. Bion-10 post-flight operations time schedule. The speed and efficiency of the search and recovery operation were remarkable. The capsule had to be removed by helicopter from the original landing site, which was in a forest. The tent was erected primarily to facilitate the medical check of the two monkeys. The autonomous experiment CLOUD was handed over to its principal investigator (who travelled with the recovery team) within four hours of landing. Biobox and the autonomous experiments were returned to Moslab 16.5 hours after landing. The KNA facilities were returned following a different schedule, which included an overnight stop at Kustanaj.

Figure 3-12. A Bion capsule after landing. The photograph shows Bion-11, which was very similar to Bion-10. Note the KNA containers on top of the capsule and the open parachute hatch. (Photo provided by Yu. Evstratov).



3.6 Post-flight Operations

3.6.1 Landing and payload recovery No ESA personnel were involved in the recovery of the Bion-10 payload (not until the recovery of Foton-11 in 1997 did ESA obtain permission to travel to the landing site). Figure 3-11 shows a reconstruction of the landing and post-landing events, based on data provided by IBMP. Additional information can be found in Section 7.6 (recovery of the exposure experiments) and in Section 6.5.3 (recovery of Biobox).

3.6.2 Post-flight activities at Moslab Biobox and the autonomous experiments were returned to Moslab within 17 hours after landing (see Section 6.5.3). To understand the



thermal anomaly Biobox had been suffering during the last part of the flight, the flight facility was immediately subjected to a series of verification tests at Moslab. No functional flaw was detected however (see Section 6.6.3).

The data download of BIOBOX and the ALGAE facility was performed at Moslab, that of FLIES and CLOUD was done next door in Building 4.

In most experiments, the sample materials (some still within, others removed from the experiment hardware) were returned as soon as possible by the investigators to their home institutes for the post-flight analysis (Figure 3-13).

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- The Buran was the counterpart of the US Space Shuttle, with comparable capabilities. After one unmanned flight in 1988, in which the pilot-less vehicle made a successful automatic landing (much to the astonishment of US experts) the programme was scrapped because of the lack of funding.
- [2] Also known as TsUP-Rokot. It is directly associated with a Moscow ground station designated as OKIK-14.
- [3] ESA's current Moscow Office (also known as the Permanent Mission) was inaugurated in 1993, i.e. after Bion-10. Moslab preceded the Permanent Mission by one year.

Figure 3-13. Transport phases of the experiments after landing.

4. Orbit Analysis

4.1 Orbit Description

4.1.1 X-Y-Z frame of reference

The most popular way to specify a satellite orbit is to use orbital elements, which refer the orbit to a frame of reference defined with respect to the stars. The commonly used frame of reference has the X- and Y-axes in the Earth's equatorial plane with the Z-axis pointing north along the Earth's spin axis. The X-axis points toward the Sun's position at the Spring equinox. This point is almost "fixed" in the inertial space, as it moves along the equator with a rate of about 0.8 arcmin per year. The Y-axis completes the right-handed orthogonal set (Figure 4-1).

4.1.2 The six classical orbital elements

The orbit of a satellite around the Earth can be regarded as an ellipse, with one of its two foci at the centre of the Earth. In Figure 4-1 the six classical or Keplerian orbital elements, which define an elliptical orbit, are shown. The ellipse lies on a plane (the orbital plane) which intersects the Earth's equatorial plane at two points called the nodes. The ascending node is the point on the equator where the spacecraft crosses from the southern hemisphere into the northern hemisphere. Two angles are needed to define the orbital plane. The right ascension of the ascending node Ω gives the angular



displacement between the X axis and the ascending node. The inclination of the orbit *i* is the angle between the two planes. The orientation of the ellipse within its plane is defined by a third element, the argument of perigee ω_p . This is the angle between the straight line joining the nodes and the vector **e** which defines the direction pointing to

Figure 4-1. X-Y-Z frame with the six classical orbital elements.

Table 4-1		
Symbol	Element	Value for Bion-10
Ω	Right ascension of the ascending node (angle)	250.4°205.2°
i	Orbital inclination (angle)	62.8°
ω_{p}	Argument of perigee (angle)	105.6°
e	Eccentricity (magnitude of an a-dimensional vector)	0.01
a	Semi-major axis (length)	6680 km
Ψ	True anomaly (angle)	0 - 360°

Table 4-1. The six classicalorbital elements.

perigee. The shape of the elliptical orbit is defined by its eccentricity e (e = |e|), and its size is defined by the semi-major axis *a*. The sixth element, the angle Ψ , referred to as true anomaly, defines the position of the spacecraft on the ellipse.

In terms of the classical orbital elements, the orbit of Bion-10 was as indicated in Table 4-1. The orbit inclination, 62.8°, was identical to the geographical latitude of the launch site Plesétsk. It corresponds to the lowest possible inclination when launching from this site and also the most economical inclination because of the maximum boost provided by the Earth's rotation. The value of the argument of perigee, 105.6°, indicates that Bion-10 flew at a low altitude at high geographical latitudes, i.e. over the northern hemisphere. The value of the eccentricity, 0.01, indicates that the orbit shape was almost circular (e = 0 for a true circular orbit). The right ascension of the ascending node started at 250.4°, decreasing by 3.9° per day due to the effect of nodal regression (which will be explained in Section 4.2.1). No fixed value can be specified for the true anomaly, as it varied from 0 to 360° during each orbit.

4.1.3 Other parameters used to describe the spacecraft orbit

To define the size and shape of the orbital ellipse, the perigee (lowest altitude attained in orbit) and apogee (highest altitude attained in orbit) are often given instead of the semimajor axis and the eccentricity. The apogee and perigee heights can be simply derived from these two orbital parameters as follows, where R_{Earth} refers to the Earth's equatorial radius (6378.14 km):

perigee height =
$$a(1-e) - R_{Earth}$$
 (4-1)

apogee height =
$$a(1+e) - R_{Earth}$$
 (4-2)

In addition, the orbital period P is often used to specify the orbit. If perturbations due to the Earth's oblateness are ignored, the nodal orbital period (i.e. the time [in minutes] from one ascending node to the next) depends only on the semi-major axis as follows:

$$P = 2\pi \sqrt{\frac{a^3}{\mu}} \approx 84.489 \cdot \left(\frac{a}{R_{Earth}}\right)^{\frac{3}{2}}$$
(4-3)

where μ is the Earth's gravitational parameter. For a circular orbit, the velocity of the satellite [in km/s] depends only on the length of the semi-major axis, i.e. the orbit radius *r*, as follows:

$$v = \sqrt{\frac{\mu}{r}} \approx 7.90537 \cdot \sqrt{\frac{R_{Earth}}{r}}$$
(4-4)

When the orbital parameters of different Bion and Foton flights are compared (Table 4-2), a consistent pattern emerges. These spacecraft fly typically in a low Earth-orbit (LEO) with a 62.8° inclination at approximately 300 km altitude. The orbit is near-circular. Each orbital

Table 4-2. Orbital parameters (inclination, period, perigee and apogee) of Bion and Foton missions with ESA payloads onboard. The parameters are nearly identical from mission to mission, except for Bion-9 which was launched in a different orbit with higher inclination required by particular radiation experiments. Sources: TRW Space Log Vols. 32, 33, 34 and (Foton-12) Space News, Satellite Digest-327.

Table 4-2.

Mission	International	Inclination	Period	Perigee	Apogee
	Designation	(degs)	(minutes)	(km)	(km)
Bion-8	1987 083A	62.8	90.5	214	382
Bion-9	1989 075A	82.3	89.2	207	267
Bion-10	1992 095A	62.8	90.3	215	364
Foton-7	1991 070A	62.8	90.4	220	404
Foton-8	1992 065A	62.8	90.2	225	372
Foton-9	1994 033A	62.8	90.3	228	385
Foton-10	1995 006A	62.8	90.3	217	363
Foton-11	1997 060A	62.8	90.4	232	385
Foton-12	1999 048A	62.8	90.5	217	384


Figure 4-2. Regression of the ascending node. The plane of the spacecraft orbit rotates with respect to the Sun. The Earth's rotation is not shown in the figure because it is unrelated to the nodal regression.

Figure 4-3. Accumulated nodal regression during the flight of Bion-10. The initial value of Ω was 250.4°. This value changed at a rate of -3.9° per day. By the end of the flight Ω was 205.2°.

loop is completed in about 90 minutes and the average speed of the satellite in orbit is close to 7.7 km/s.

4.2 Time Evolution of Orbital Parameters

4.2.1 Nodal regression

As is the case for any satellite in a low Earthorbit, the orbital trajectory of Bion-10 was not a static and closed ellipse. Perturbation forces caused the satellite orbit to vary with the time. The two main sources of perturbation are the Earth's gravitational field and air drag. Owing to the polar flattening (or the equatorial bulge), the Earth's mass distribution is not uniform and this affects the right ascension of the ascending node Ω . The uneven mass distribution of the Earth produces a torque on the satellite, which rotates the angular momentum vector. For an orbit with an inclination of less than 90° (for Bion-10 it was 62.8°) the orbital plane rotates in a westerly







Figure 4-4. Apogee height evolution during the Bion-10 flight (reworked by Kayser Italia from basic data provided by Perry's Kettering Group). Launch time: 29 December 1992 13:30 UTC (16:30 h Moscow time).

Figure 4-5. Perigee height evolution during the Bion-10 flight (reworked by Kayser Italia from basic data provided by Perry's Kettering Group). Launch time: 29 December 1992 13:30 UTC (16:30 h Moscow time).

Table 4-3. Apogee and perigee values for Bion-10 as provided by different sources. Legend: ESOC = European Space Operations Centre; PKG Kayser Italia = data provided by PKG and reworked by Kayser Italia; PKG = Perry's Kettering Group; Molniya = Molniya Space Consultancy; TRW = TRW Space Log. direction. In Figure 4-2 the regression of the node is shown.

The rate of nodal regression per orbit in radians per revolution is given to a first approximation by the following formula:

$$\Delta \Omega = \frac{3 \pi J_2 R_{Earth}^2 \cos i}{a^2 (1 - e^2)^2} \tag{4-1}$$

where J_2 is a constant (1082.78 x10⁻⁶) which reflects the mass distribution of the Earth. For the orbit of Bion-10, with its inclination of 62.8°, $\Delta\Omega$ is about -3.9° per day. The minus sign means that the orbital plane moves to the west. Over the 11.6-day flight of Bion-10, the accumulated nodal regression was approximately -45° (11.6 x -3.9°). In other words, between launch and landing the orbital plane of Bion-10 had rotated substantially with respect to the Sun (Figure 4-3). As will be explained in Section 4.5, the effect of nodal regression was to over-expose the spacecraft to solar light during the second half of the flight.

4.2.2 Loss of altitude

The effects of the air drag are mainly concentrated within a small orbital arc around perigee. Loss of energy near the perigee means that the altitude at the next apogee is lower than the previous one, so that the orbit progressively evolves towards a circular shape (i.e. both *a* and *e* decrease with the time). This
5) explains the paradox that the altitude at perigee decreases very slowly with respect to variations in apogee, despite the fact that almost all the drag is concentrated around r perigee (Figures 4-4 and 4-5).

For the Bion-Foton orbit, the combined variations in the heights at perigee and apogee due to aerodynamic effects causes the semimajor axis to decrease by about 450 m/day (28 m/rev or 6.3 km per 14 days). Thus, during a two-week mission the apogee may be expected to decrease from its initial value by about 12.6 km. On the other hand, the height at perigee is only slightly affected, and changes by no more than 1 to 2 km over a two-week period. It must be pointed out that these variation rates are strongly affected by

Table 4-3.

Parameter	KB Photon	ESOC	PKG Kayser Italia	PKG	Molniya	TRW
Apogee (km)	397	385373	374360	387372	376	364
Perigee (km)	226	219217	217215	219217	218	215

changes in atmospheric density owing to solar activity. The 11-year solar cycle can make the atmospheric density vary by a factor of 10 in the region around 300 km altitude, with major consequences for the evolution of the orbit. To a first approximation, the change per revolution in the semi-major axis is proportional to the atmospheric density. The Bion-10 mission took place near to a maximum in the solar cycle and so the decrease in altitude was greater than average. The perigee decreased by about 2.5 km in less than 12 days of flight, while the apogee decreased by about 15 km during the same time (Figures 4-4 and 4-5).

One should also note that differences can be found in the values of the semi-major axis, the perigee, the apogee and other parameters provided by different sources (Table 4-3), even when they are based on the same raw radar observation data. In fact, the precise definition of some key orbital parameters depends on the orbit propagation model, for instance, it depends on the consideration of osculating vs. mean elements or on the mathematical models of the Earth's gravitational field and of the atmosphere.

4.3 **Ground Tracking**

4.3.1 Longitude shift between orbits

An important characteristic of the orbit from the point of view of a satellite user is the evolution with time of its projection point on the Earth's surface (the sub-satellite point). An indication of the variations in the sub-satellite point is given by the shift in the ascending node at each new revolution. Between two successive orbits, the ascending node changes relative to an observer on Earth by a longitude

Table 4-4.



angle $\Delta \phi$, owing to the combined effects of regression of the orbital plane (as shown in Figure 4-2) and the Earth's rotation. After completing one orbit, the spacecraft does not return to its exact point of departure and furthermore, the Earth has rotated somewhat about its own axis during the one-and-a-half hours it took the spacecraft to complete the orbit. This shift is given to a first approximation by the following expression:

$$\Delta \phi = \Delta \Omega + \Delta \phi_R$$

where
$$\Delta \phi_R = -\frac{2 \pi P}{T_E}$$
(4-6)

where T_E is the period of revolution of the Earth (24 hours), P is the satellite's orbital period (~ 90 min) and $\Delta\Omega$ is the regression of the ascending node per revolution. The term $\Delta \phi_{R}$ takes account of the motion of the satellite relative to the Earth. Recalling that the nodal regression is negative (i.e. it moves from East to West), the plus sign in the formula means that the Earth's rotation

Figure 4-6. Bion-10's first three ground tracks after launch. Consecutive orbits have ground tracks that are shifted by 22.9° to the West. This effect is due to a combination of the Earth's rotation and nodal regression.

Table 4-4. Longitudinal shift
between successive orbits at the
start and end of the flight of
Bion-10.

Parameter	Start of flight	End of flight	Units
Nodal regression $\Delta\Omega$	-0.244	-0.245	degs/rev
Longitude shift by Earth rotation $\Delta \phi_{R}$	-22.66	-22.64	degs/rev
Total shift $\Delta \phi (= \Delta \Omega + \Delta \phi_R)$	-22.904	-22.885	degs/rev

augments the longitudinal shift due to nodal regression. The values of these parameters for the Bion-10 mission are given in Table 4-4.

A longitudinal shift of -22.9° means a westerly longitude separation between two consecutive ground tracks of 22.9°. This appears as a displacement of about 2560 km at the equator as the spacecraft passes from one orbit to the next (Figure 4-6).

4.3.2 Daily longitude shift and delay time

From a user point of view, it is convenient to arrange that the satellite flies over the same site (e.g. a ground station) at the same time each day. This type of orbit is called a 24-hour Earth-synchronous orbit. This is to say that the sub-satellite point follows a ground track identical to that of a previous orbit one day earlier. This requires that an integer number (n) of orbits later, the value of $\Delta \phi$ accumulated over a complete day will be equal to 2π . For Bion-10, the number of orbits per day ranged from 15.9292 to 15.9468, and so the orbit was not 24-hour Earth-synchronous. In fact, for an observer on Earth, the daily shift in the ascending node [in degrees] was:

$$\Delta \Omega_{observer} = 16 \cdot \Delta \phi - 2\pi \approx 6.4 \tag{4-7}$$



This implies that the day after a given orbit has taken place, an observer at the ascending node will see the satellite equator crossing at about 700 km to the west, but with a delay of about 5 minutes, as simply explained by the following expression:

$$Node_{delay} = 16 \cdot P - 24 \cdot 60 \approx 5 \tag{4-8}$$

4.3.3 Orbit numbering conventions

It is important to note that KB Photon has adopted an orbit numbering method that differs from the NORAD convention used in the West. For the Russians, orbit No. 1 simply starts at launch. For NORAD, orbit No. 1 is defined as the loop that begins at the first ascending node (i.e. the first time the spacecraft crosses the equator going north). Thus, NORAD starts counting from 1 when the Bion-Foton spacecraft has already completed three quarters of an orbital loop. (The latter is referred to by NORAD as orbit No. 0). Accordingly, the serial numbers assigned by NORAD are usually one unit smaller than the corresponding KB Photon reference. For some reason, the orbit numbers assigned to Bion-10 by NORAD were not one, but three units smaller than those assigned by KB Photon. This must have been due to an error in the NORAD timelines, as Bion-10's orbit No. 0 was tagged at 19:05 h Moscow time, whereas the launch actually took place at 16:30 h Moscow time.

4.3.4 Launch window

Bion and Foton capsules are launched from Cosmodrome Plesétsk in Northern Russia located at longitude 40.4° E, latitude 62.8° N. The typical launch sequence for Bion-Foton missions exploits the 62.8º latitude of the launch site in Plesétsk. A local horizontal and eastward injection is performed in a typical launch with an inclination of 62.8°. The gain in the burn-out velocity due to the Earth rotational velocity is quite low (0.2 km/s) because of the high latitude. On the other hand, the launch site allows an orbit with an inclination equal to the launch site latitude to be achieved with the minimum amount of energy, i.e. thrust in the direction of motion is used only to expand the orbit.

The launch window is the time at which the launch site on the surface of the Earth passes

Figure 4-7. The launch window of Bion-10. At 16:30 h local time, the Earth's rotation had placed Plesétsk in the correct position for launching the spacecraft into the pre-selected orbital plane. The shaded area indicates the part of the Earth in shadow. through the chosen orbital plane. Since for Bion and Foton missions the orbit inclination is equal to the launch site latitude, only one daily launch window exists for a particular orbital plane in inertial space (Figure 4-7). In this case the orientation of the orbital plane depends on the time of the launch and is given by the following relationship:

$$\Omega = LST + \pi/2 \qquad ($$

where Ω is the right ascension of ascending node (RAAN) and LST is the local sidereal time.

For practical reasons, Bion and Foton launches do not generally take place at the following times:

- The cosmodrome is closed on Saturday and Sunday. Since pre-launch activities for scientific missions need an uninterrupted lead time of more than one day, closure during the weekend means that no launch can take place on a Monday.
- During the winter, weather conditions at the landing area (located on the border between Russia and Kazakhstan) are unfavourable for search and recovery operations.

4.3.5 Re-entry and landing

The track of the last orbit of Bion-10 in free flight is shown in Figure 4-8. The retrorocket





was ignited at 06:44:33 (Moscow time) on 10 January 1993, when the satellite was over South Africa. The braking manoeuvre changes only the position of the spacecraft within the orbital plane, so that the ground track remains quite constant during the re-entry, apart from a small Eastward shift due to the reduction in orbital period. Bion-10 was originally planned to land on 12 January. Because of the increase in onboard temperature, the flight controllers decided to land the spacecraft two days ahead of schedule. Assuming that on 10 January the same landing protocol was applied as that prepared for 12 January, one would expect the re-entry ground track to intersect the landing area 2 x 6.4° further East (Section 4.3.2), resulting in a touchdown 12.8° East of the originally planned landing site. This estimate is in good agreement with the geographical location of the planned and the actual landing site. The nominal landing place of the spacecraft was the vicinity of Kustanaj (63º E, 53º N), while the actual landing took place 100 km north of Karagandá (73º E, 56º N). (Figure 4-8).

The landing is usually scheduled to take place shortly after sunrise, to take advantage of the calm air conditions in the upper layers of the atmosphere and to ensure that search and recovery operations can be completed in daylight. Figure 4-9 shows the intersection of the Bion-10 trajectory with the landing area two hours after sunrise. Figure 4-8. Bion-10's last orbit. The retrorocket was fired about half an hour before touchdown, when the satellite was over South Africa. After the braking manoeuvre, the spacecraft still moves in the original orbit plane and so, the ground track on re-entry is similar to the one obtained for the orbital free flight. Touch down was at 07:16 h (Moscow time). The actual landing site, near Karagandá, and the originally landing site at Kustanaj, were both within the boundaries of the vast landing area used for Bion-Foton missions.

Figure 4-9. The time of landing is usually scheduled to occur within two hours after sunrise, to take advantage of calm air conditions in the upper atmospheric layer. This was also the case for Bion-10. Table 4-5. Visibility features at perigee for Bion-10. Perigee 217 km, inclination 62.8°, grazing angle 5°.

Table 4-6. Visibility features at apogee for Bion-10. Apogee 360 km, inclination 62.8°, grazing angle 5°.

Figure 4-10 (left). Sub-satellite point and visibility circle.

Figure 4-11 (right). Visibility circles.



4.4 Visibility and Earth Coverage

4.4.1 Visibility features

To illustrate how the visibility of the satellite is influenced by the geometry of the relation between the Earth and the satellite orbit, a simple model assuming a spherical Earth is used. At a given instant, the satellite is visible from all points on the Earth's surface within a circle centred on the sub-satellite point, and having a diameter increasing with the satellite altitude (Figure 4-10).

An observer on the perimeter of the visibility circle sees the satellite on the horizon, i.e. at an elevation angle ε equal to zero. The distance from this observer to the satellite is called the slant range; it is the distance from the satellite to the surface of the Earth along a tangent to the surface of the Earth. The circumference of the visibility circle defines an angle λ_0 , subtended at the centre of the Earth, within which all points on the surface of the Earth within the visibility of the satellite will lie. The angle λ_0 and the slant range *s* vary with the satellite altitude *h*, according to the following expressions:

$$s = \sqrt{h^2 + 2R_E h}$$

 $\lambda_0 = \arccos\left(\frac{R_E}{R_E + h}\right)$

Visibility features	Typical value
Slant range s	1680 km
Angular coverage angle λ_0	14.7 degs
Angular coverage angle λ_5	10.5 degs
Entry point distance d	1210 km
Coverage arc	1175 km
Coverage area A	$4 \times 10^{6} \text{ km}^{2}$
Max observation duration Tmax	5.5 min

Table 4-6.

(4-10)

(4-11)

Visibility features	Typical value
Slant range s	2170 km
Angular coverage angle λ_0	18.8 degs
Angular coverage angle λ_5	14.4 degs
Entry point distance d	1690 km
Coverage arc	1600 km
Coverage area A	8×10 ⁶ km ²
Max. observation duration Tmax	7.5 min

From the geometry involved, it is clear that the angle λ (which defines a circle on the surface of the Earth from which the satellite is seen with the same elevation ε on the horizon), decreases as e increases. Since radio communications at low angles of elevation incur considerable signal attenuation owing to atmospheric affects, the surface coverage is



Date	Orbit No.	Session timing	Session duration (n
30 Dec	15	13:40 - 13:44	4
	16	15:13 - 15:18 16:59 16:45 orror!	5
	17	18.15 - 18.18	3
	19	19:46 - 19:51	5
31 Dec	28	09:07 - 09:10; 09:07 - 09:12; 09:11 - 09:17	3, 5, 6
	29	10:40 - 10:45; 10:45 - 10:50	5, 5
	30	12:13 - 12:18	5
	31	13:39 - 13:44; 13:47 - 13:52	5, 5
	32	16:45 - 16:50	5
	34	18:20 - 18:25	5
01 Jan	44	09:07 - 09:10; 09:07 - 09:12; 09:11 - 09:17	3, 5, 6
02 Jan	60	09:21 - 09:26; 09:22 - 09:28; 09:27 - 09:28	5, 6, 1
	61	10:56 - 11:01	5
	62	12:29 - 12:34	5
	63 64	14:02 - 14:07 15:28 - 15:33	5
	65	17:01 - 17:06	5
03 Jan	76	09:28 - 09:33; 09:29 - 09:35; 09:34 - 09:39	5, 6, 5
	77	11:03 - 11:08	5
	78	12:36 - 12:41	5
	79	14:09 - 14:14	5
	80 81	15:35 - 15:40 17:05 - 17:13	5 8
04 Jan	92	09:32 - 09:45: 09:37 - 09:42	13.5
	93	11:06 - 11:10	4
	94	12:39 - 12:44	5
	95	14:12 - 14:16	4
	96 97	15:37 - 15:43 17:10 - 17:15	6 5
)1	17.10 - 17.15	5
05 Jan	108	09:36 - 09:42; 09:42 - 09:46; 09:45 - 09:50	6, 4, 5
	109	11:14 - 11:19 12:47 12:52	5
	110	12.47 - 12.32 14.09 - 14.14	5
	112	15:46 - 15:51	5
	113	17:19 - 17:24	5
06 Jan	124	09:43 - 09:48; 09:47 - 09:52	5, 5
	125	11:16 - 11:21	5
	126	12:49 - 12:54	5
	127	14:15 - 14:19 15:48 - 15:53	4
	128	17:21 - 17:25	4
07 Jan	140	09:47 - 09:51; 09:48 - 09:52; 09:52 - 09:57	4, 4, 5
	141	11:29 - 11:36	7
	142	13:02 - 13:07	5
	143	14:29 - 14:34	5
	144	10:01 - 10:00 17:25 - 17:30: 17:36 - 17:30	5 3
	175	17.23 - 17.30, 17.30 - 17.37	5,5

Table 4-7.

nin)

Table 4-7. Telemetry session start and end times for the Bion-10 mission as announced by the TsUP during the mission (see also Chapter 8). The orbit numbers refer to the KB Photon standard. The date and time refer to Moscow local time.

Date	Visible orbits	Visible	Non-visible
	(orbit no.)	(total)	(total)
30 December	12, 13, 14, 15, 16, 17, 18, 19	8	8
31 December	28, 29, 30, 31, 32, 33, 34	7	9
01 January	44, no further data	?	?
02 January	60, 61, 62, 63, 64, 65	6	10
03 January	76, 77, 78, 79, 80, 81	6	10
04 January	92, 93, 94, 95, 96, 97	6	10
05 January	108, 109, 110, 111, 112, 113	6	10
06 January	124, 125, 126, 127, 128, 129	6	10
07 January	140, 141, 142, 143, 144, 145	6	10

Table 4-8. Visible orbits versusnon-visible orbits. Dataextracted from Table 4-7.

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typically restricted to the region in which the satellite elevation above the horizon is greater than 5° (Figure 4-11).

In general, the observation time for a given visibility window for a particular ground station is given by:

$$T = \frac{P}{\pi} \cdot \arccos\left(\frac{\cos\lambda_{\max}}{\cos\lambda_{\min}}\right)$$
(4-12)

where λ_{max} and λ_{min} represent the maximum and minimum angles subtended at the centre of the Earth during the passage of the satellite. It must be noted that the duration of the visibility window depends strongly on λ_{max} which in turn is linked to the minimum elevation angle for which the telemetry link is active. For example, if a ground station is sited on a mountain top, ε_{\min} could be 2°, offering 15% more viewing time compared to the case with $\lambda_{\min} = 5^{\circ}$. The maximum observation time is obtained with an overhead pass ($\lambda_{\max} = \lambda_0$ and $\lambda_{\min} = 0$) and it is simply given by:

$$T_{\max} = P \cdot \frac{\lambda_0}{\pi} \tag{4-13}$$

Assuming for Bion-10 a grazing angle of 5° , the visibility parameters take the values given in Tables 4-5 and 4-6.

4.4.2 Duration of individual telemetry sessions

In conclusion, the maximum uninterrupted contact time between the Bion-10 spacecraft and any individual ground station cannot have been more than 7.5 minutes. The actual start and end times of Bion-10's telemetry sessions were announced by TsUP during the mission (for more details: see Chapter 8). This allows us to check the estimated values of T_{max} (Table 4-5 and 4-6) against the actual contact times (Table 4-7). Indeed, all telemetry sessions were terminated within 7.5 minutes, with two exceptions: the last session on



Figure 4–12. The sub-satellite area and the ground station area of Bion-10. The subsatellite area spanned 62.8°N to 62.8°S, the ground station area was restricted to 43-64°N and 23-167°E.

Date	first TM session start time	last TM session end time
	(Moscow time)	(Moscow time)
30 December	08:55	19:51
31 December	09:07	18:25
01 January	09:07	not known
02 January	09:21	17:06
03 January	09:29	17:13
04 January	09:32	17:15
05 January	09:42	17:24
06 January	09:43	17:25
07 January	09:48	17:39

Table 4-9. Synchronisation of telemetry sessions with the Moscow working day. Data extracted from Table 4-7.

Figure 4-13. The spacecraft passes over the territory of the CIS in daytime. In the morning (Moscow local time was 09:00 h) the spacecraft started to cross the far eastern part of Russia. By the end of the working day, it was still over Russia. At night, the spacecraft was never in touch with the ground stations. Legend: A, B, C, orbital orientation at the beginning of flight. D, E, F, orbital orientation at the end of flight.

3 January lasted 8 minutes and the first session on 4 January lasted 13 minutes. The announced timing of the latter session must have contained an error (cf. the third session on 30 December). **4.4.3 Daily pattern of the telemetry sessions** Table 4-7 shows more characteristics of the telemetry schedules of Bion-10. From 2 January onwards, a regular daily pattern was established in which telemetry sessions took





Figure 4-14. Sun illumination conditions for a satellite.

place during six consecutive orbits, alternating with ten consecutive orbits that were devoid of telemetry contact (Table 4-8). Each day started with a double or triple telemetry session (Table 4-7), presumably to download accumulated data stored onboard during the ten previous orbits when no contact was made with the ground stations. Two, even three neighbouring ground stations may have been involved in these multiple sessions. In Table 4-7 the daily delay time effect, as explained in Section 4.3.2 can be noted. For example, the telemetry session schedules for 2-7 January were nearly identical, except that every following day the entire schedule was delayed by a few minutes.

4.4.4 Synchronisation of telemetry sessions with Moscow office hours

Another conclusion that can be drawn from the schedule of telemetry sessions (Table 4-7) is that the spacecraft was intended to make contact with the ground exclusively during the normal working hours in Moscow. As shown in Table 4-9, Bion-10's earliest telemetry session that we know of started at 08:55 h Moscow time, while the latest telemetry session ended at 19:51 h Moscow time. Usually, the spacecraft dumped its data between 09:00 h and 17:30 h Moscow time. This indicates that the TsUP in Moscow would receive fresh data from the spacecraft exclusively during the normal office hours.

Following completion of the mission, the entry and exit times of the telemetry sessions (Table 4-7) were correlated with the satellite's ground track, which had been reconstructed by Kayser Italia, using data provided by NORAD. From this it was deduced that Bion-10's telemetry sessions were almost exclusively linked to ground stations located between 43-64° N and 23-167° E. In other words, the ground stations used for Bion-10 were all located within the territory of the CIS (Figure 4-12).



Figure 4-15. Sun's motion for one orbit, represented in a spacecraft-centred celestial coordinates. The conclusion that Bion-10's ground stations were located on CIS territory may not come as a surprise. More puzzling however is the question how telemetry synchronisation with the TsUP working day was obtained. If the ground station network had been distributed all over the globe, this would have been fairly easy. Contact could have been established at any time of the day, and ground stations could be switched on and off at will to fit the Moscow office hours. However, the actual ground station area used for Bion-10 was not worldwide, but confined to a restricted zone within the total sub-satellite area (Figure 4-12). Apparently, the spacecraft flew only over this restricted area in synchronism with the Moscow working day. This effect could have been achieved by selecting the time of launch during the day. This assumption is confirmed when a reconstruction is made of Bion-10's orbital trajectory versus its position towards the ground station area (Figure 4-13). From a knowledge of the orbital orientation of Bion-10, it is clear that around 09:00 h in Moscow, Bion-10 was flying over Russian territory (Figure 4-13(A) and (D)). By the end of the day the spacecraft moved out of sight of the ground station zone (Figure 4-13 (B) and (E)). Although the orientation of the Bion-10 orbit shifted throughout the mission over 45° by nodal regression, the situations at the start and the end of the mission were still largely comparable (compare Figures 4-13(A), (B) and (C) with (D), (E) and (F)). It is obvious that there was no need to de-activate the ground stations outside the Moscow working day, as there simply were no passes over Russian territory during the Moscow night (Figure 4-13(C) and (F)). Furthermore, as the spacecraft never passed over any ground station at night, no night shift was required to man ground stations, irrespective of their geographical location or time zone.



4.5. Eclipse Conditions

In Figure 4-14, the regions of space illuminated by the Sun and those in eclipse are shown for a generic orbit. In the situation depicted by the figure, the orbit has a portion that lies in the conical shadow region called the umbra, where the Sun is completely hidden by the Earth. The direction of the shadow cone is defined by the line joining the centres of the Sun and the Earth. Outside the umbra is the penumbra, where a portion of the Sun is blocked from view and consequently the illumination of the satellite is reduced, but it is not zero. It is clear that the portion of the orbit during which the spacecraft is in eclipse is an arc of the ellipse that lies on the section of the shadow cone given by its intersection with the orbital plane. The length of this arc (which may be zero) depends on both the orientation of the orbital plane with respect to the Sun-Earth direction and the satellite's height above the Earth's surface.

For a given size and shape of the orbit, as determined by its semi-major axis *a* and eccentricity *e*, the orbital plane can be chosen (inclination *i* and right ascension Ω) to obtain either eclipse-free orbits (maximum illumination by the Sun) or the maximum eclipse arc (maximum shadowing). For the Bion-Foton missions, the inclination *i* fixed at 62.8°, the amount of shadowing per orbit depends solely on the right ascension Ω . As explained above in Section 4.2.1, the value of

Figure 4-16. The eclipse history of Bion-10, as reconstructed by ESOC. The horizontal axis denotes time (in days) and the vertical axis the timespan of a single orbit. The vertical stripes, which together form a cigarshaped pattern, each represent the fraction of the 1.5-hour orbit that was spent in darkness. At the beginning of the flight, this fraction spanned 0.40 to 0.98 h, i.e. 0.58 h or approximately 35 minutes per orbit. This value declined gradually until, on 7 January, the spacecraft flew out of eclipse, i.e. no part of its orbit was in darkness.

Figure 4-17. The Bion-10 eclipse history, as provided by KB Photon/IBMP. The onboard temperature history is shown by solid bullets. The horizontal axis denotes time in hours, and the left-hand vertical axis temperature in °C. The eclipse profile is indicated by the dotted line; the right-hand vertical axis represents the time (in minutes) per orbit spent in darkness.



 Ω was not constant during the flight of Bion-10 as the orbital plane rotated by 45° during the course of the mission due to nodal regression. Because of this regression, and because of the Earth's motion on its orbit around the Sun (about 1° per day during the mission), the Bion-10 orbit gradually moved out of the umbra. **4.5.1 Eclipse orbit fraction (EOF)** A more effective representation that aids understanding of the geometry involved in the eclipse phenomenon uses a celestial sphere reference, centred on the spacecraft (Figure 4-15). The Sun ephemerides and the satellite orbit plane orientation are assumed constant during a single orbit. And since the typical



Figure 4-18. Comparison of the actual eclipse profile, the optimum profile corresponding to a launch delay of about 4 h 44 min. and an intermediate profile corresponding to a launch delay of 3 h. The effects are due to the different orientations of the orbit with respect to the Sun. Values of the right ascension of the first ascending node, determined by simulations performed by Kayser Italia for the actual. optimum and intermediate cases, are 250.4°, 321.4° and 295.4° respectively.

eccentricity Bion-Foton orbit is 0.01, the orbit can be considered circular, neglecting minor effects due to perigee orientation.

The celestial coordinate system is centred on the spacecraft. The Z-axis lies on the normal to the orbital plane, defined as the equator, and the X-axis is given by the satellite-Earth direction, which points to the nadir on the equator. In this coordinate system, the Earth disc is shown by the shaded circle (Figure 4-15). Because the X-axis is fixed on the surface of the Earth, the coordinate system will rotate by one turn around the polar axis (orbit pole) for each satellite revolution. Thus, assuming the Sun to be fixed at a certain position on its ecliptic plane during an orbit, it will appear to rotate once per orbit about the orbit pole, describing a parallel circle. An eclipse will occur whenever this solar circle lies on the Earth disc. The duration of the eclipse is given by the fraction of orbital period equal to the fraction of this circle that lies on the Earth disc. As may be deduced from Figure 4-15, the value of this fraction (usually called the eclipse orbit fraction) depends on the Sun angle σ with respect to the orbit plane and the angular radius of the Earth disk ρ , according to the following formula given by simple considerations of spherical geometry:

$$EOF = \frac{1}{\pi} \cdot \arccos\left(\frac{\cos\rho}{\cos\sigma}\right)$$
(4-14)

The Earth's angular radius ρ is simply related to the orbit altitude *h* by the relation:

$$\rho = \arcsin\left(\frac{R_E}{R_E + h}\right) \tag{4-15}$$

On the other hand, many parameters contribute to the value of the Sun angle σ . In fact, it is a complex function of the Sun's ascension α and the declination δ ; in practice this means the day of the year, and the parameters of the orbital plane of the satellite, i.e. the inclination *i* and the right ascension of the ascending node Ω . From considerations given in Figure 4-15, an eclipse-free orbit occurs if the following condition is met:

$$|\sigma| \ge \rho$$

with (4-16)
 $|\sigma|, \rho \le \frac{\pi}{2}$

The Sun angle s ranges from a maximum of $(i + \varepsilon)$ to a minimum value of $-(i+\varepsilon)$, where ε is the ecliptic obliquity ($\varepsilon = 23.4^{\circ}$). In the case of a typical Bion-Foton orbit (inclination $i = 62.8^{\circ}$ and mean orbit altitude h = 300 km), we obtain a maximum for the Sun angle $\sigma_{max} \approx 86.3^{\circ}$. The Earth angular radius ρ becomes about 72.7°, so that if σ can vary from 72.7° to 86.3° (and from -72.7° to -86.3°) no eclipse will occur. On the other hand, the maximum EOF is obtained for $\sigma = 0$, and so for the assumed orbit:

$$EOF_{max} = \frac{\rho}{\pi} = \frac{72.7}{180} = 0.4 \tag{4-17}$$

corresponding to a maximum eclipse duration of about 36 min, assuming an orbital period of about 90 min $(0.4 \times 90 \text{ min} = 36 \text{ min})$.

4.5.2 Eclipse profile

Depending on the season of the year and, in particular, the time of launch, the value of the Sun angle σ can vary over a period of two weeks from zero (EOF = 0.4) to a maximum value (EOF = 0). This happened during the 11.6-day flight of Bion-10. After the flight, the eclipse history of Bion-10 was evaluated at ESOC (by G. Janin) using orbit data provided by NORAD (Figure 4-16). A very similar eclipse history was later provided by KB Photon (Figure 4-17). In the latter diagram, the EOF is plotted together with the on-board temperature. The effect of the absence of shadowing on the spacecraft temperature is obvious. As soon as the spacecraft flew out of eclipse, the temperature could no longer be regulated. It should be noted that by choosing a different launch time, the overheating of the payload in the last part of the flight could have been avoided. By software simulation, it is possible to identify which launch time would give the best EOF profile (whereby the maximum eclipse condition is maintained during the whole flight). In Figure 4-18, various EOF profiles are given. If the launch had been delayed by 4 hours 44 minutes,



Attitude of the Bion-10 orbit w.r.t the sun at the beginning of the flight



Attitude of the Bion-10 orbit w.r.t the sun at the end of the flight

Figure 4-19. Out of eclipse

corresponding to a right ascension of the first ascending node of 321.4°, the optimum Sun radiation shielding would have been obtained for Bion-10. In this case the satellites would have been in the umbra region for about 40% of the flight time for the whole duration of the flight, allowing the spacecraft thermal control to maintain the environmental payload temperature within the nominal limits.

4.6. Conclusions

On 7 January 1993, after nine days of flight, the Bion-10 spacecraft flew out of eclipse (Figure 4-16). From this event onwards, a progressive temperature rise was set in motion (Figure 4-17) which threatened the payload such that the spacecraft had to make an emergency landing two days before the planned date. The question of why the spacecraft flew out of eclipse was never satisfactorily answered, neither by the mission management nor elsewhere. At that time, it seemed that flying out of eclipse could easily have been avoided – simply by launching at another hour of the day, or by shifting the launch date to another part of the year (Section 4.5). However, based on the orbital analysis performed by Kayser Italia, under contract to ESA, and complementary information, we have come to the conclusion that, given the circumstances, the mission authorities probably had no other choice than to allow the spacecraft to fly out of eclipse, and accept the calculated risk of overheating. The following list of questions and answers explains how we arrived at this conclusion.

- **Q.** How do we know that the uncontrolled temperature rise on board was due to the out-of-eclipse-conditions?
- A. This was officially confirmed by the Bion-10 mission management (RKA, TsSKB, IBMP) in a post-flight meeting with ESA in Moscow in 1993.
- **Q.** *When is an orbiting object out of eclipse?*
- A. This is illustrated in Figure 4-19. When the plane of the orbit is perpendicular to the Sun-Earth axis, the spacecraft is continuously illuminated by the Sun. This is called the out-of-eclipse condition. For full details, see Section 4.5.
- **Q.** When did Bion-10 go out of eclipse?
- A. Bion-10 went out of eclipse during the second half of the flight. Bion-10's eclipse history was, after flight, independently calculated by ESOC (using Two-Line Element data supplied by NORAD) and by TsSKB (using Russian satellite tracking data). The results were near identical (Figures 4-16 and 4-17). Initially, there were between 34 and 35 minutes of shadow during each 90-minute orbit. In the course of the flight, the number of shadow minutes per orbit fell gradually to zero.
- **Q**. Why could the favourable flight conditions of the first half of the flight not be maintained during the second half?
- A. That was because the orbital plane of any Earth-orbiting object drifts slowly but continually with respect to the Sun owing

to nodal regression (Section 4.2), together with the Earth's motion on its orbit around the Sun. These combined effects caused the orbit orientation of the Bion-10 with respect the Sun to move slowly from a favourable to an unfavourable condition.

- **Q.** Could flying out of eclipse have been avoided ?
- A. Yes, by choosing a different orientation of the orbital plane at the beginning of the flight. However, with parameters like the location of the launch site (Plesétsk) and launch direction (due East) fixed, the initial orientation depended solely on the time of launch. If Bion-10 had been launched some hours later, the spacecraft would not have gone out of eclipse (Section 4.5).
- **Q.** If so, why was Bion-10 not launched some hours later?
- A. If it were, the spacecraft would not have flown over Russia during daylight.
- **Q.** So what? What would have gone wrong if the spacecraft had not flown over *Russia during daylight*?
- A. In that case, the spacecraft could not have landed on Russian soil in daylight. Landing in daylight (preferably in the early morning) is a necessary condition for speedy recovery, which is especially important for a biosatellite (see Section 4.3.5). In addition, flying over Russian territory in daylight means that the ground station network and the flight control

centre (all on Russian territory) can be operated during normal working day hours, without night shifts (Section 4.4.4).

- Q. If launched in another period of the year, the Bion-10 spacecraft could have been injected into an orbit that allowed for crossing the Russian territory during daylight without flying out of eclipse (Section 4.5). So, why was the launch of Bion-10 not moved to a more favourable calendar date?
- **A.** Bion-10 was ready for flight at the very end of 1992, some months later than planned. The date of launch could not be postponed any further, into 1993, for a variety of reasons. Firstly, the budget for Bion-10 was only secured for a launch in 1992. A second reason was the arrival of the severe Russian winter. As a rule, Bion capsules are not flown during the winter because the snow and cold in the landing area hinders the recovery operations. A third, less compelling reason was that Bion-10 was dedicated to the 1992 International Space Year. In short: a further delay, into a period with superior eclipse conditions, was financially, operationally and politically unacceptable.

Main source for this chapter: Kayser Italia's Bion-10 Mission Analysis, (KI-FOPS-SYS-TN-007), 1997.

This chapter was checked by Dr. Luciano Anselmo of CNR-CNUCE.

5. The Experiment Environment

5.1 Radiation

5.1.1 Radiation from space

According to the documentation provided by KB Photon [1, 2], the radiation dose rate at the outer surface of the spacecraft can be as high as 510 mGy/d (51 rad/d), not taking into account the rare contributions of solar flares. The reported worst-case value for space radiation entering into the capsule is 550 μ Gy/d (0.055 rad/d, in the absence of solar flares (Figure 5-1(A)). Judged by these figures, the dosage of the cosmic rays is attenuated by three orders of magnitude by the combined shielding effects of the capsule wall, the battery pack and the service module (Figure 5-1(B)). The density of the capsule wall varies from 9.14 g/cm^2 , on the bottom, to 2.11g/cm² on the top (Figure 5-1(C)). The spacecraft re-enters the atmosphere with its base in front, which is why this part is endowed with a heavier skin than the top. It can be stated that, irrespective of its exact location, the payload inside the capsule is always shielded by at least 2.11g/cm², this figure representing the thinnest part of the capsule wall. No figures have been published about the shielding densities of the battery pack and the service module.

5.1.2 On-board radiation source

Besides being exposed to radiation coming from space, the payload was constantly irradiated by an on-board gamma-ray source. ESA learned about the existence of this source during the Bion-9 mission (1989) and obtained details about the source's location and characteristics after Bion-10. The radionuclide used for the gamma source is the caesium-137 isotope (Table 5-1). The source, which is located off-set in the bottom of the re-entry capsule, is part of an altimeter that plays an



important role during landing by triggering small braking rockets to cushion the impact of the landing. The rockets are fired when the capsule, hanging from its parachute, has descended to an altitude of less than a few

Figure 5-1. Summary of the radiation data taken from the hardware design specifications of KB Photon. Table 5-1. Characteristics of thegamma-ray source on boardBion.

Radionuclide	caesium-137
Half-time	30.2 years
Energy	0.661 MeV
Dose rate at 50 cm distance	1040 µGy/day
	(=104 mrad/day)
Intensity (calculated)	3.04 mCurie

metres above the ground. Similar retrorockets, fitted to the parachute shrouds, are used to cushion the impact shock of the well-known manned Soyuz capsules returning from Mir and the ISS.

According to KB Photon [1, 2], the payload receives 1040 μ Gy/d at a distance of 500 mm from the gamma-ray source. Since the internal diameter of the spacecraft capsule measures about 2 m, and because radiation decreases with the square of the distance, the approximate dose rate in relation to the position of the payload (assuming no shielding in between the payload and the source) is that given in Figure 5-1(D).

Figure 5-2. Duration of the exposure to space radiation and to gamma-rays from the onboard source. Exposure to the onboard radiation source was for a longer duration than the exposure to space radiation.

High up in the capsule, at a distance of 2 m from the source, the expected dose is approximately 65 μ Gy/d plus up to 550 μ Gy/d due to space radiation. In this location, radiation



from space will be the predominant contributor to the daily dose. If the capsule is fully packed with experiments, the gamma-ray source will be well shielded, reducing its contribution even more.

By contrast, experiments installed low down in the capsule may be exposed to more than 1040 μ Gy/d from the gamma-ray source, plus up to 550 μ Gy/d due to space radiation. Note that in this position, the experiments are bound to receive a higher daily dose from the onboard source than from the cosmic rays! In retrospect, such may have been the case for the experiments ALGAE, CLOUD, FLIES, WOLFFIA and parts of DOSICOS and SEEDS, which were all mounted near the floor of the Bion-10 capsule (see Figures 2-7 and 2-8).

The contribution of the gamma source to the total dose must be even stronger than to the daily dose, because the payload is exposed to the gamma source *for a longer time* than to space radiation: exposure to the gamma source starts two days before launch, when the payload is integrated with the capsule, and ends some time after landing, when the payload is removed from the capsule. On Bion-10, the exposure to space radiation lasted for 11.6 days, whereas the exposure to the gamma-ray source spanned 13.6 days (see Figure 5-2).

Results obtained from ESA's DOSICOS experiments flown on Bion-9 [3, 4] confirmed that the onboard source can contribute significantly to the total absorbed dose. The DOSICOS detectors located at the outer surface of the capsule (out of reach of the gamma-source and thus exposed to space radiation only) recorded a total dose 2 mGy behind a shielding of 2.0 g/cm². Inside the capsule, behind shielding of more than 2.1 g/cm², the total dose varied from 2.5 to 5.8 mGy depending on the position of the detector. This suggests that the contribution of the on-board source to the dose was at least 20% in the first case, and at least 65% in the second case.

This disconcerting result was, however, not prominently reproduced on Bion-10. Outside the capsule, 250μ Gy/d was recorded behind

2.0 g cm², whereas inside the capsule, where the shielding was more than 2.1 g/cm², the highest measured value was just a little more at 263 μ Gy/d (Chapter 9-4, DOSICOS). This would suggest that on Bion-9, the DOSICOS detectors were placed in closer proximity to the source than on Bion-10.

For more radiation measurements made on Bion-10, see the discussion of the DOSICOS, SEEDS and WOLFFIA experiments in Chapter 9 and references [5] and [6].

5.1.3 Conclusions

Depending on their position in the capsule, experiment packages in the Bion spacecraft may be exposed to gamma radiation from the onboard source to the extent that, dose-wise, more radiation is received from the source than from space. This conclusion is based on the figures provided by KB Photon, and is corroborated by the results from ESA's DOSICOS experiment on Bion-9. There is, however, no evidence that outside-exposed experiment packages were affected by the gamma-ray source.

What effect the gamma-ray source may have on the outcome of the biological experiments onboard is uncertain. It is known that some components of the radiation spectrum are more harmful than others. Heavy charged particles (HZE) from space are particularly dangerous because their energy deposition is extremely dense, both spatially and in time. In contrast, gamma radiation scores low since its interaction with matter is smoothly spread in space and time. The conclusion can be drawn that the on-board source can easily influence the total dose readings inside the capsule, whereas its biological effects are supposedly limited. Still, it would be premature to deny that the gamma source could affect the results of the experiments onboard, because biological effects can reportedly be induced by chronic, low-dose gamma radiation [7].

5.2 Microgravity

No recorded microgravity data are known from Bion. Recently, data have been published

about the microacceleration levels as measured on board the very similar Foton capsules. From these, it appears that values better than 10^{-4} g are obtained along all three orthogonal axes.

5.3 Atmosphere Inside the Capsule

Pressure in the spacecraft	710 - 767 mm Hg
O ₂ partial pressure	143 - 188 mm Hg
\tilde{CO}_2 partial pressure	0.5 - 1.1 mm Hg
Relative humidity	40 - 70%

It should be noted that only the fruit flies of the FLIES and CLOUD experiments were in direct contact with this environment. All other experiments flown jointly by ESA and IBMP were encased in hermetically sealed containers.

5.4 Temperatures

5.4.1 Temperature inside the capsule

The temperature history, recorded inside the capsule, is illustrated in Figure 4-17. The temperature was maintained at a stable level until 7 January, when the spacecraft went out of eclipse (See Sections 4-5 and 4-6). Values

Figure 5-3. The ambient temperature in the capsule was recorded in five different locations by ESA (shown as red dots). In addition, KB Photon provided ESA with recordings of temperatures taken at three locations, shown by the blue dots.



over 30°C were attained at the end of flight. The temperature was measured in many different locations in the capsule (Figure 5-3). Profiles similar to that shown in Figure 4-17 were recorded in each spot, but owing to local differences (thermal gradients) the values differed by several degrees. The highest temperatures were recorded by the ALGAE experiment (the peak value was 32.5°C) and the T0 sensor, attached externally to Biobox, which reached 34.5°C. In contrast, the sensors attached to the inlet and outlet of the airconditioner (shown in Figure 5-3 as the cooling/drying device) never saw values higher than 28.4°C at the inlet and 27.8°C at the outlet. The relatively cool environment around the airconditioning unit and the hotter environment elsewhere in the capsule suggest that the air circulation inside the capsule may not have been sufficient.

5.4.2 Temperature inside Biobox

The temperatures recorded inside ESA's Biobox incubator are given in Section 6-6.

5.4.3 Temperature outside the capsule

The temperatures recorded in the KNA exposure facilities, at the outer surface of the capsule, are given in Section 7-8.

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6. Biobox

6.1 The Birth of Biobox

After the satisfactory results of the Bion-8 and Bion-9 missions, interest was raised within ESA to use the Bion capsules more intensively. Nevertheless, these plans could not be realised without first removing an obstacle, namely the temperature variations in the Bion spacecraft. The temperature range specified for Bion was 18-28°C, which, at first sight, was adequate to keep the typical Bion passengers such as monkeys, rats, plants, insects and fish in good health. However, as recordings from Bion-8 and Bion-9 indicated, the spacecraft temperature was not stabilised to a tightly controlled nominal value and it underwent excursions over the entire temperature range 18-28°C, even exceeding these limits on some occasions [1].

To ensure that the control experiments on ground would be subject to the same temperature history as the flight experiments, IBMP adopted a standard policy of conducting the reference experiments after flight, once all recorded flight data had been reviewed and analysed. For this purpose, a mock-up of the Bion capsule was used at IBMP Plánernaya in which the in-flight thermal environment as recorded, including all of its anomalies, deviations and fluctuations, was carefully reproduced. By itself, this was a sound approach. However, from Biorack's maiden flight in 1985 onwards [2], the ESA life science community had become accustomed to a different philosophy whereby the 1g ground reference experiments were conducted simultaneously with the flight experiments. This method was deemed scientifically superior, because it guaranteed that the reference data were never biased by timerelated variations in the biological specimens

such as biorhythms, maturation, ageing and so on.

On the Bion-8 and Bion-9 missions, ESA had attempted to introduce the *in synchrony* philosophy, but the results were disappointing. Reproduction of the capsule's in-flight temperature synchronously on-ground requires that all pertinent spacecraft data be available on-ground in real time. Experience with Bion-8 and Bion-9 taught that the telemetry was never available in real time; the delay time between in-flight monitoring and the receipt of data at IBMP could span more than half a day. It was therefore not possible to conduct the ground control experiments synchronously with the in-flight experiments [1].

A second-best option was then chosen by ESA, in which the time frame of the control experiments was delayed with respect to the flight experiments by one complete day (This was termed near-synchrony). Already a compromise, this solution was further frustrated when it was discovered that the Bion-9 telemetry did not reflect the true temperature of the ESA-provided flight experiments. The reason for this (which, by the way, was only fully understood on Bion-10 when a similar mismatch was found, see Section 5.4.1) was the existence of thermal gradients within the capsule. The single on-board temperature sensor, which supplied the telemetry data, had been placed, by an unfortunate coincidence, in a hot spot and its readings were not representative of the typical experiment conditions on board [1].

As a result of these considerations, the idea of developing an incubator specifically for Bion matured. Such an incubator would guarantee that the experiment temperatures were fully controlled throughout the flight. Moreover, by making the incubator pre-programmable, the temperature profile of the flight experiments would be predictable. That being the case, the in-flight temperature could be replicated on ground without the need for telemetry, and the control experiments could be conducted in synchrony without undue additional effort.

Another advantage offered by the envisaged incubator was the possibility of delivering controlled temperatures outside the 18-28°C band. Unexpected reactions had been observed in mammalian cell cultures under weightlessness. (By 1990, one of the mostacclaimed space biological experiments was the suppression of lymphocyte activation in microgravity reported by Cogoli et al. [2, 3]; another was the decrease in *c*-fos induction in epithelial cells reported by De Groot et al. [4].) Culturing of mammalian cells requires an ambient temperature that is stabilised at around 37°C. Flying an incubator on Bion would make these experiments feasible. The availability of pre-set temperature control spawned new ideas directed at the other end of the temperature scale. If the incubator could lower its temperature down at the end of flight, thus acting as a refrigerator, this would prevent chemically-fixed samples from decaying.

Finally, providing the incubator with its own centrifuge would allow reference experiments to be conducted under a constant acceleration of 1g in flight, to simulate exposure to

terrestrial gravity. (These would be additional to the reference experiments conducted on ground). By doing so, the quality of the experiments could considerably be enhanced. The in-flight 1g control was, and still is, considered to be the best reference available, because the control samples experience an environment that is exactly the same as their counterparts exposed to microgravity (including launch vibrations, space radiation, landing shocks) except for one single factor: gravity [3]. IBMP had occasionally flown such centrifuges on Bion from 1975 onwards, carrying fruit flies, turtles and even rats. ESA's Biorack [2, 3, 5, 6] (first flown in 1985) was also equipped with 1g centrifuges but, unfortunately, these could not be used to their full efficacy. Ideally, the reference samples should start spinning immediately after launch. In the busy agenda of the Space Shuttle crew, the Biorack centrifuges never obtained first priority so that the reference samples were usually waiting for many hours in unwanted weightlessness at the beginning of the flight. During this period, irreversible biological changes could be introduced [3, 6, 7]. By contrast, the incubator for Bion, which would be fully automated, would never be troubled by crew-time limitations. It could potentially out-perform Biorack by delivering 1g to the reference samples immediately upon orbital injection. A name for the Bioncompatible space incubator was quickly found: Biobox, a working title that emphasised its kinship with Biorack, was soon adopted as the official name.

Table 6-1.

Company

Dornier GmbH (D) Kayser Italia S.r.l. (I) Verhaert Design & Development N.V. (B) Carrar S.A. (F) Centrum voor Constructie en Mechatronica b.v. (NL)

Responsibility

prime contractor; thermal design facility controller; EGSE chassis; incubator box; MGSE centrifuge; centrifuge controller experiment housing & electronics; experiment units*

* Functionally, the experiment units were an integral part of Biobox but contractually they were not. The units were developed in conjunction with the investigators and financed by the national funding agencies SRON, Prodex and (to a lesser extent) CNES.

Table 6-1. Members of theBiobox consortium.



Figure 6-1. Section of an automatic experiment unit as used in Biobox. Biobox carried 30 such units. Drawing: CCM.

6.2 The Biobox Industrial Consortium

Biobox was designed and manufactured under contract to ESA by a European consortium consisting of Dornier (now part of Astrium GmbH) as the prime contractor, supported by Kayser Italia (I), Verhaert (B), Carrar (F; a company that ceased to exist in 1995) and CCM (NL) (Table 6-1).

6.3 Biobox Described

6.3.1 The experiment units

To explain the functional anatomy of Biobox, the facility is best described from inside out, starting with the internal subsystems and ending with the external chassis.

The innermost parts of Biobox were represented by the experiment units. Biobox carried thirty of them. In each unit, one or two biological cultures could be grown. The units were of uniform size $(2 \times 4 \times 8 \text{ cm})$ and conformed to a standard set earlier by ESA's Biorack Project. This worked in the interest of the investigators since the Biobox experiment units were interchangeable with those of Biorack. Within this small volume, one or two 1-ml cell culture compartments were accommodated plus six 1-ml fluid storage membranes. Upon issuing an electrical command signal, the storage vessels could be emptied one-by-one into the culture compartments, replacing the fluids that had been standing there before. In a typical experiment, a sequence of culture medium



Figure 6-2. A CIS (= Cells In Space) container. Biobox carried three CIS containers. Each held eight experiment units (two stacks of four) and an ECU (electrical control unit). The ECU is located right, behind the CIS container lid, in the compartment decorated with the CCM label. The space underneath the ECU was used as a gas reservoir. Before flight, the CIS containers were filled with 5% CO_2 in air. CO_2 exchange with the cell cultures was possible through the experiment unit's permeable membrane (see Figure 6-1). Photo: CCM.

Figure 6-3. An ESA Type-I container as used in Biobox. Biobox carried eight Type-I containers on its centrifuge; six were assigned to experiments and two were occupied by ECUs (electrical control units). Left: to provide CO₂ to the cell cultures, the Type-I containers could be fitted with a gas reservoir (the bulged structure on top) and a gas valve. Right: the experiment units were mechanically and electrically attached to the Type-I container lid. Photo: CCM.





Figure 6-4. Layout of the Biobox experiment platform on Bion-10. The experiments BONES, MARROW and OBLAST consisted of seven experiment units in the CIS containers plus two on the centrifuge. FIBRO was composed of three units, with none on the centrifuge. The configuration on ground was identical, except for the fact that no experiments were assigned to the centrifuge.

Figure 6-5. Biobox, with the lid removed, factory-fresh in June 1992. The three CIS containers and the centrifuge were mounted on a platform inside the incubator box. The picture shows Biobox on its handling frame, the two hand-grips fitted to a rectangular support structure. The handling frame was removed before flight. Photo: Dornier. replenishments was concluded by supplying a fixing solution to chemically preserve the biological cultures. Figure 6-1 illustrates the internal layout of a Biobox experiment unit.

The Biobox experiment units were based on a proven CCM design, flown since 1985 on Biorack [8] and several sounding rockets [9]. The main differences between these earlier units (the "first generation") and the ones used in Biobox (the "second generation") were the



latter's improved capability for sterilisation, optimised biocompatibility and the optional provision of gas exchange between the cell culture compartments and the experiment unit's external atmosphere.

6.3.2 Experiment housing and electronics Of the thirty experiment units, twenty-four (three groups of eight) were housed in aluminum cases known as CIS containers (Figures 6-2 and 6-4). Like the experiment units themselves, the CIS containers were derived from a proven CCM design used for ESA's sounding rocket programme [9]. The six remaining experiment units were individually encased in Biorack Type I containers (Figure 6-3).

The fluid replacement sequences inside the experiment units were managed by electrical control units (ECUs). Each CIS container was equipped with one internal ECU (serving eight experiment units, Figure 6-2) while the six Type I containers were fitted with two external ECUs, each ECU serving three experiment units. The experiment timelines (i.e. the timing of the fluid replacements, with individual scenarios for each experiment unit) were programmed inside the ECUs: the commands were issued through a command line. In addition, the ECUs were able to monitor experiment-related data such as plunger command acknowledgements and temperature and pressure histories.

6.3.3 The centrifuge

The six experiment units that were housed in the six Type I containers were placed on a centrifuge through which they were subjected to an acceleration of 1g during flight. This set of six served as an in-flight control to the 24 microgravity-exposed experiment units in the CIS containers. The centrifuge was derived from the device developed and supplied by Dornier for Biorack, with Biobox-specific adaptations made by Carrar. The modifications included the addition of a cable to lock the rotor during launch and landing, and the capability to carry two ECUs. The latter was realised at the expense of two experiment positions on the rotor. So, although the Biobox centrifuge did carry eight Type I containers -







Table 6-2.

mass (gross weight) mass (experiment platform empty) outer dimensions inner dimensions (incubator box) incubator temperature setpoint range power consumption (average) power consumption (peak) energy consumption (17d) thermal control system just like the original Biorack centrifuges – two of these were not occupied by experiments, but by electronics. The two Type I containers holding the ECUs were known as SCBs (switch and control boxes), see Figure 6-4.

6.3.4 The experiment platform

The three CIS containers and the centrifuge were mounted on a rectangular support plate called the experiment platform, which served as both a mechanical interface and an electrical interface for power and data transfer. The positions of the CIS containers, the centrifuge and the experiment units are shown in Figure 6-4. An electrical harness, routed underneath the plate, provided 18 digital command lines, 18 digital status lines and 28 analog input lines.

6.3.5 The incubator

To provide a controlled temperature environment to the experiments, the platform with its CIS containers and centrifuge was placed inside a thermal box (Figure 6-5). As *pars pro toto*, this box was strictly speaking the "Biobox". The floor, the four side walls and the removable lid were all built of insulating Rohacell panels. One side wall of the box was fitted with a thermal assembly consisting of ten Peltier elements (Figure 6-6). To smooth thermal inhomogeneities in the incubator volume, fans were placed in the box to create a forced flow of air.

The incubator was equipped with ten thermal sensors to measure the temperature inside and outside the box. In addition, the experiment temperatures were measured by a separate set of eight thermal sensors. Figure 6-6. Biobox: the thermal assembly. Two sets of five Peltier coolers were externally fitted to the side of the incubator box, covered by two rectangular structures. Air was sucked in from above, and expelled through six pipes blowing down. Photo: Dornier.

Figure 6-7. Biobox: the facility controller and the centrifuge controller. The facility controller is the big rectangular box with the connector panel on top. The centrifuge controller is the slim black box at left. Photo: Dornier.

Figure 6-8. The Biobox fleet-ofthree consisted of two flightcertified models (including one spare) and one non-flyable qualification model. The latter was used for flight-synchronous reference experiments on ground. Photo: Dornier.

42.5 kg 30.4 kg 70 × 44 × 34 cm (l × w × h) 37.5 × 36.5 × 14 cm (l × w × h) 14-37°C 50 W 125 W 20 kWh 10 Peltier elements

Table 6-2. Characteristics ofBiobox.

Figure 6-9. Integration of **Biobox with the Bion-10** spacecraft in Plesétsk, in the evening of 27 December 1992. Crouched inside the capsule, ESA contractor Mr. L. Pieroni (Kayser Italia), verifying the mating of the satellite interface connectors. The carton box in his hand contained silicon glue, used for the fixation of the connector screws. It was the last verification step before Biobox was switched on. At a later stage in the satellite integration sequence, the monkey Ivasha was seated in the space where Mr. Pieroni had been working. Note the proximity of the Biobox thermal assembly to the foam padding over the parachute compartment at right. Poor air circulation around the thermal assembly is thought to have contributed to Biobox's inability to cool properly down upon telecommand TK2. Also note the satellite interfaces with the KNA containers (see Chapter 7): two (dissimilar) circular seats for the KNAs are visible above the open hatch; the thin white cables hanging down on the left and the right were used to hold the KNA lids. Photo: D. Zolesi.



6.3.6 The facility controller

The ECUs and the Peltier devices were controlled centrally by the facility controller (FC), an electronics module located beside the incubator box (Figure 6-7). All time-lined switching functions were programmed in the FC. These included temperature set-points, temperature change rates, ECU activation commands and commands to the centrifuge (start, stop, lock and unlock the rotor).

The FC also contained a removable 512-Kbyte RAM card onto which the housekeeping data of Biobox was recorded, allowing the engineers to reconstruct the performance of Biobox after flight. The card data contained more detail than the telemetry transmitted to ground during flight. Data were collected every 5 minutes.

The FC also functioned as the electrical interface with the Bion spacecraft. Biobox received electrical power and telecommands through a connector panel mounted on top of the FC. Telemetry data sent out by Biobox was also channelled through this panel (Figure 6-7).

6.3.7 The centrifuge controller

The centrifuge controller was an electronics module which, like the experiment-controlling ECUs, was slaved to the FC (Figure 6-7). Under software control, the centrifuge speed was maintained at a steady, pre-selected rotation value.

6.3.8 The chassis

The incubator and its contents, the facility controller and the centrifuge controller were mounted together onto a ladder-like support structure, consisting of two longitudinal beams interconnected by struts. This chassis also served as Biobox's mechanical interface with the spacecraft (Figures 6-6 and 6-7).

6.3.9 Characteristics of Biobox

The requirements as laid down by ESA in the Biobox design specification were all met, with one notable exception. To guarantee optimal sample preservation, the incubator had to be able to cool down to 4°C. However, the lowest temperature that Biobox, as flown on Bion-10, could reach was only 14°C (Table 6-2).

Table 6-3. ESA's Biobox Team.

Table 6-3.

ESTEC

Wim Jansen Jean-Christophe Ronnet Arthur Knell María López Cotarelo René Demets Fred Franz

Industry:

Wolfgang Scheller, Dornier Matthias Fehrenbach, Dornier Luca Pieroni, Kayser Italia Fabrizio Carrai, Kayser Italia Valfredo Zolesi, Kayser Italia

Hans Leysten, CCM Jan Rietema, CCM Antoon Koppen, CCM (Bion-10 Mission Manager) (System Engineer) (Operations Manager) (Quality Control) (Scientific Coordinator) (Technical Support)

(Project Leader)
(Electrical and Operations Engineer)
(Electronics Hardware Engineer)
(Software Engineer)
(Project Leader electrical onboard and ground support systems)
(Experiment Integration Supervisor)
(Experiment Integration Electrical Engineer)
(Experiment Integration Mechanical Engineer)

A set of three Bioboxes was manufactured (Figure 6-8), comprising a flight model (FM), a flight spare (FS) and a qualification and test model (QM, Figure 6-8). The FM was selected for flight on Bion-10. However, when a bent connector pin was discovered in one of the CIS containers shortly before flight, the FM was grounded and the FS was flown instead. The QM was used for the synchronised 1g reference experiments on-ground. In the following, the FM and QM are referred to as Biobox/flight and Biobox/ ground respectively. Programmed to execute the same experiment protocol, Biobox/flight and Biobox/ground were expected to perform identically.

6.4 Biobox Planned Performance

6.4.1 Telecommands

Biobox was originally meant to fly in a fully automatic mode, with mission-specific parameters like temperature setpoints, centrifuge spin rate and experiment activation timelines pre-programmed in its memories. The start up of the facility was even envisaged to occur automatically upon orbital injection, being triggered by its own microgravity sensor, and it would shut down part of its subsystems as soon as the experiments were finished. Although these features *were* eventually implemented, the Russian flight controllers objected to a complex payload that could not be

Table 6-4.

Experiment units integrated with Biobox	start:	25 Dec	17:45 MT = L -94h, 45 min
	finish:	26 Dec	07:15 MT = L -81h, 15 min
Biobox handed-over to IBMP for transpor	t to Plesétsk	26 Dec	09:00 MT = L -79h, 30 min
Biobox integrated with spacecraft		27 Dec	19:00 MT = L -45h, 30 min
Launch (L)		29 Dec	16:30 MT = L

Table 6-4. Pre-launch timeline ofBiobox.

controlled from ground. Therefore, a limited set of overriding telecommands was added. These were designated TK1, TK2, TK3 and TK4, in addition to the mechanical start command called K0.

K0: START

The start command K0 was automatically presented to Biobox once the spacecraft was separated from the launcher, about 9 minutes after lift-off. The effects of K0 were several: the Biobox centrifuge rotor was unlocked and started to spin-up, the experiment command sequences were initiated and the incubator temperature was raised to activate the biological cultures. By the same K0 command, other instruments on board the Bion capsule were also kicked into action.

TK1: START (back-up)

TK1 was a redundant start command, uplinked from ground 12 minutes after lift-off (i.e. 3 minutes after K0). The effects of TK1 and K0 were identical.

TK2: PREPARE FOR LANDING

When the experiment command sequences were finished and all cell cultures had been safely fixed, TK2 was uplinked to prepare Biobox for landing. TK2 would trigger several events at once: the incubator temperature was lowered to a level conducive to sample preservation, and the centrifuge was stopped and locked.

TK3: POWER SAVING MODE

Using telecommand TK3, the power consumption could be set to a minimum, in the case that an emergency occurred during orbital flight (and it did on Bion-10). By sending TK3, Biobox's most power-hungry subsystems, like the Peltier devices and the centrifuge controller (but not the ECUs, the data acquisition etc.), were switched off. In other words, the facility remained active, but the experiment environment was no longer controlled. Recovery from TK3 was possible by sending a new TK1 command.

TK4: SEND DATA

Issuing TK4, requested Biobox to provide

housekeeping data. TK4 was, in practice, never uplinked from ground. It was, however, automatically issued by the spacecraft on-board computer. TK4 was sent every hour, resulting in Biobox delivering fresh data 24 times per day for storage in the spacecraft computer. How the stored data were relayed to ground is explained in Chapter 8.

All telecommands sent to Biobox/flight, were also sent to Biobox/ground through its electronic ground support equipment, but with a time delay of 75 minutes.

6.4.2 Incubator temperature profile

The incubator temperature profile was programmed to enter the following steady states consecutively: SLEEP, RUN and STORE. The program commenced as soon as the cell cultures had been loaded in the incubator, more than three days before launch. During the SLEEP state, the temperature was held at -20°C to arrest cell growth and development prior to the flight. SLEEP was maintained until the spacecraft was in orbit. During the RUN state, the temperature setpoint was 36.5°C, this value having been derived from the optimal temperature for mammalian cell growth (37°C) minus a 0.5°C safety margin to cater for temperature overshoot. Biobox was programmed to make a controlled transition from the SLEEP to the RUN state in just over three hours at a rate of 5°C per hour. When all experiment command sequences were finished, and all cell cultures had been fixed (nine days after launch), the incubator was switched from the RUN state into the STORE state (14°C), again at a controlled rate of 5°C per hour (to be completed in slightly more than four hours). The incubator temperature of 14°C during the STORE state, the lowest temperature attainable in Biobox, was meant to hinder the decay of the fixed cultures.

6.4.3 Centrifuge speed

The selected centrifuge speed for Biobox/flight was 112 rpm. At this rate of rotation, the biological cultures (located at 72 mm distance from the rotor axis) were subjected to 1.0g.



Biobox/ground was operated vertically, tilted by 90° , to ensure that the Earth's gravity vector would intersect the cell cultures at the same angle as the 1*g* vector on the centrifuge in Biobox/ flight. In this position, the spin axis of the centrifuge was horizontal. Hence, no experiments were carried out using the centrifuge on ground, the experiment positions on the rotor being occupied by dummies (Figure 6-4).

6.4.4 Experiment command timelines

Each of the four experiments in Biobox featured different experiment protocols with, accordingly, different timelines. In addition, different experiment units within one experiment often followed different timelines. For further details, see Chapter 9.

6.5 **Biobox Operations**

6.5.1 Pre-flight operations

The two Biobox models were prepared in Moslab by a team whose fourteen members were drawn from ESTEC and industry (Table 6-3). Biobox was ready for transportation to the launch site three days before launch, and was integrated into the spacecraft less than 48 h before lift-off (Table 6-4).

6.5.2 Operations during flight

During flight, Moslab was staffed by Wim Jansen and Arthur Knell, who followed the Biobox/flight telemetry as transmitted from the TsUP to Moslab (full details of this are given in Chapter 8) and who maintained surveillance of the Biobox/ground.

As planned, Biobox/flight was switched from the SLEEP state (20°C) into the RUN state (36.5°C) by the dual commands K0 and TK1 upon injection into orbit. When, nine days into flight, TK2 was sent to initialise the STORE state, the centrifuge stopped turning and was locked as planned. However, TK2 did not cause Biobox to reach the planned STORE temperature (14°C). Noting this anomaly, the flight controllers decided to quieten Biobox by issuing the emergency command TK3, as further detailed in Section 6.6. The exact timing of the telecommands sent to Biobox is given in Table 6-5.

6.5.3 Post-flight operations

The Bion-10 capsule landed in the early morning, two hours after sunrise, in a snowcovered forest. The helicopters of the recovery team arrived almost immediately at the landing spot. The capsule was sling-carried by a Mil Mi-8 helicopter to a place near the forest where Figure 6-10. Biobox experiment temperatures (in °C) during flight. The temperatures were measured in four locations by four pairs of thermistors: one pair in each CIS container, one pair on the centrifuge. Each of the four plotted lines represents the average reading from a sensor pair. The temperature increase on 7, 8 and 9 January was due to an in-flight thermal anomaly in the spacecraft (see Section 6.6).

Figure 6-11. Biobox experiment temperatures (in °C) on ground (reference experiments). The temperatures were measured in three locations by three pairs of thermistors: one pair in each CIS container. Each of the three plotted lines represents the average value from a pair. No experiments were allocated to the centrifuge on ground. The temperature rise on 8 and 9 January was a deliberate attempt to re-create on ground the in-flight thermal anomaly (see Figure 6-10).

Table 6-5.

command	effect	date	Moscow time	elapsed time since lift-off
K0 (nominal)	start	29 December	16:38:47	8 min, 47 sec
TK1 (nominal)	start	29 December	16:41:30	11 min, 30 sec
TK2 (nominal)	stop	07 January	17:36:10	9 days, 1 h, 6 min, 10 sec
TK3 (off-nominal)	power saving	08 January	16:07:30	9 days, 23 h, 22 min, 30 sec
TK4 (nominal)	delivery of data	every hour through	hout the whole mission	

Table 6-5. Commands sent to Biobox.

Table 6-6.

10 January 1993			
Biobox power switched from main to aux		06:48	R - 28 min
Landing		07:16	R
Biobox de-integrated from spacecraft	start:	10:30	R + 3 h, 14 min
	finish:	11:00	R + 3 h, 44 min
Biobox power off/on		10:54	R + 3 h, 38 min
Biobox departure from landing site		14:00	R + 6 h, 44 min
Biobox arrival in Moscow		22:30	R + 15 h, 14 min
Biobox back in Moslab		23:50	R + 16 h, 34 min
11 January 1993			
De-integration of experiment containers	start:	12:00	R + 28 h, 44 min
	finish:	18:50	R + 35 h, 34 min
(time rendered in Moscow Time = $GMT + 3$)	h)		

Table 6-6. Post-landing timelineof Biobox.

a tent was erected to protect the satellite and the recovery team against the biting cold (-22°C). The inside of the tent was heated (14-17°C) and here Biobox was dismounted from the capsule, reconnected to its transport batteries and loaded into its thermally controlled transport container by IBMP's Aleksey Alpatov and Murad Tairbekov.

In the afternoon, Biobox was transported by helicopter to the civil airport at Karagandá, where it was loaded into an Antonov An-12 cargo plane. At 22:30 h, the plane landed at Chkálovskaya (also known as Shchëlkovo), a military airbase in the northern outskirts of Moscow. From there, Biobox was transported by bus to IBMP Plánernaya and it arrived at Moslab just before midnight (less than 17 h after landing). The following day the recorded data were downloaded and the experiment units were dismounted and handed over to the investigators. The timing of the post-flight operations (based on data taken from references [15, 16]) is given in Table 6-6.

6.6 Biobox Performance as Flown

6.6.1 Temperature profile

The first two steady states, SLEEP and RUN, were executed as planned. The transition to the third steady state, STORE (14°C), was to be triggered by telecommand TK2. When TK2 was presented, Biobox started to cool down as planned. However, having descended to 17°C, the temperature began to rise again slowly to finally settle, more than ten hours later, at 24°C. In short, Biobox failed to attain the desired STORE temperature (Figure 6-10).

The off-nominal temperature rise inside Biobox was paralleled by an unexpected temperature increase *outside* Biobox, in the Bion-10 capsule. This made TsUP suspicious that the dysfunctional Biobox was generating too much heat. Fearing that the capsule might become dangerously overheated, the controllers at TsUp decided to set the power consumption of Biobox at minimum by sending TK3. No longer controlled, the temperature in Biobox then followed the ambient temperature of the capsule, which went over 30°C for the remaining part of the flight (Figure 6-10).

6.6.2 Temperature accuracy

The experiment temperatures were measured by eight sensors: two in each of the three CIS containers, and two on the centrifuge. The resolution of the thermal sensors was 0.2°C (50°C over 256 levels).

The temperature differences across the four locations (CIS-1, -2, -3, centrifuge) remained within the specified 1°C band during each of the three steady states except for SLEEP in Biobox/flight, indicating a virtual absence of thermal gradients in the incubator. Moreover, the stability of the temperatures was very good; at each individual location the temperature was steady within 0.2°C during SLEEP and RUN (Figures 6-10 and 6-11).

Less satisfying was the realisation of the selected temperature values. There was a conspicuous mismatch between the setpoint values and the temperatures actually attained. As is apparent from Figures 6-10 and 6-11, all experiment temperatures during the RUN state were too high, with values ranging between 37.0°C and 38.0°C rather than between 36.0°C and 37.0°C.

6.6.3 Temperature rise outside Biobox

Once the power consumption of Biobox had been set to minimum by issuing TK3, the temperature in the capsule did not return to its normal level, but climbed even further, contrary to the assumption by TsUP that the temperature rise in the spacecraft was due to Biobox. This behaviour suggested that some existing thermal anomaly in the spacecraft had caused the temperature of Biobox to rise, instead of vice versa. The evidence to support this alternative explanation was later produced during tests performed after flight at Moslab. Back on ground, the cooling system of the Biobox/flight unit was certainly fully functional and the incubator was perfectly able to reach, and to stay at, the planned STORE temperature (14°C). The question of why Biobox failed to do so in flight was never

fully clarified, although indirect evidence from ground tests suggested that the performance of the Biobox heat exchangers was degraded by the combination of the high ambient temperature and the limited air circulation in the capsule (see Section 5.4.1).

The true cause of the abnormal temperature rise in the capsule *was* later identified and has been explained in Chapter 4. Launched under unfavourable orbital conditions, the spacecraft flew "out-of-eclipse" on day nine of the flight, after which it was exposed to the Sun for longer than was normal.

About one year after completion of the Bion-10 flight, an official from TsSKB declared that the mission controllers at TsUP knew right from the start that Biobox was not to blame for the thermal problems encountered in the capsule. In fact, mission control switched Biobox off because it was the only heatproducing equipment on board that could be shut down by telecommand.

6.6.4 Plunger activation

With thirty experiment units in flight plus twenty-four on ground, a total of 54 experiment units was used in the Bion-10 mission. Each unit was equipped with up to six programmable plungers, the total number of plungers being 312. Of these, two plungers got stuck halfway through their stroke, the other 310 travelled over the full path. The success rate was thus 99.4% [10]. The two failures occurred together in one unit from the experiment MARROW flight series. The cause was probably an incorrect alignment of membranes during the pre-flight integration. All 312 plungers were fired in full compliance with the protocols programmed into the ECUs.

There was, however, a divergence between the in-flight and on-ground timelines which is discussed in Section 6.6.6.

6.6.5 Centrifuge speed

From the third day in orbit, the centrifuge in Biobox/flight started to spin too fast. The nominal rate of rotation, 112 rpm, increased to a measured rate of 119 rpm. However, 119

Table 6-7. Centrifuge spin ratesand ensuing g-values.

Table 6-7.			
g-value			
1.00 g			
1.14 g			
1.33 g			
1.54 g			

Bion-10 spacecraft during the last days of the flight as monitored by the reference sensor T6. Vertical axis: temperature, horizontal axis: elapsed flight time, expressed in number of completed orbits. The four vertical dotted lines refer to the following events: 1. After 145 orbits, telecommand TK2 (Enter STORE mode) was sent to **Biobox**; 2. After 160 orbits, telecommand TK3 (Shut down) was sent to Biobox; 3. After 174 orbits, the spacecraft was commanded to undergo a re-orientation manoeuvre; 4. After 186 orbits, the capsule touched down. Note that the temperature continued to rise after 160 orbits, even though Biobox had been shut off. The re-orientation manoeuvre apparently had a beneficial effect on the

temperature, perhaps because

the re-entry capsule became shaded by the service module.

Figure 6-12 Temperature in the

rpm was also the highest value that could be monitored by the speed sensors and the real spin rate may have been greater. The anomaly could be reproduced on ground; in post-flight tests speeds of 130-140 rpm were recorded. These values may reflect the actual speed attained in flight. The impact on the *g*-value is indicated in Table 6-7.

6.6.6 Flight simulation on ground

The Biobox/ground unit was operated in Moslab with a delay of 75 minutes with respect to the Biobox/flight unit. However, due to an error on the part of the Biobox operators, the entire plunger activation timeline in Biobox/ground was shifted 3 hours and 34 minutes forwards. As a result, rather than 75 minutes later, each plunger on ground was activated more than two hours earlier than its counterpart in flight. Meanwhile, TK1 and TK2 were sent to Biobox/ground as planned, that is, 75 minutes after Biobox/flight received these commands. Upon receipt of TK2, Biobox/ground assumed the STORE state without any problems being encountered - unlike Biobox/flight. Once it was realised that the STORE state had failed in flight, attempts were made to reproduce this anomaly on ground. However, in the absence of real-time telemetry, the in-flight temperatures could not be copied with full precision (compare Figure 6-10 with Figure 6.11).



6.7 Biobox in the Press

The flight of Biobox on Bion-10, including the thermal problems which arose, was extensively covered by the media [11, 12, 13, 14]. These reports contained some inaccuracies that need to be rectified.

In Reference [11], the temperature history of Biobox became confused with that of the capsule, making the discussion incomprehensible:

"...mission controllers had deliberately raised temperatures in the capsule to 37 degrees Celsius to incubate some of the experiments...".

Of course, the temperature was not deliberately raised in the capsule, but in *Biobox*.

"Then the capsule was to be cooled down to 14 degrees Celsius."

The same misunderstanding: only Biobox, not the entire capsule, was to be cooled down.

In Reference [14] Prof. Ilyin, Head of the Bion Programme, unjustly claimed that Biobox contributed significantly to the temperature rise in the capsule:

"The situation was aggravated by the power consumption of the ESA Biobox, which was higher than anticipated...".

There is no factual evidence to support this statement. In contrast, in a post-flight debriefing in Moscow on 19-21 April 1993, called one week after Ilyin's letter was published, and attended by representatives of ESA, IBMP and KB Photon, the conclusion was drawn that the power consumption of Biobox had been within specification throughout the entire flight.

"Within 12 hours after Biobox was placed in its near-dormant state, the capsule temperature had dropped to 27.7 Celsius." Again, there is no recorded data to support this statement. On the contrary, after Biobox was placed in its near-dormant state by sending TK3, the temperature in the capsule continued to climb for more than sixteen hours (eleven orbits) afterwards (Figure 6-12).

6.8 Conclusions

A number of shortcomings came to light during the mission. The most conspicuous, that Biobox/flight did not reach 14°C in the STORE state (Section 6.6.1), was undoubtedly induced by the thermal problems in the spacecraft and cannot therefore be blamed on Biobox itself. The rumour, circulated in the press, that the thermal problems in the Bion-10 spacecraft were due to Biobox have no foundation in fact whatsoever, as explained above (Section 6.7).

Other anomalies were:

- 1. the inaccurate realisation of the selected experiment temperature levels (Section 6.6.2);
- 2. the centrifuge speed overshoot (Section 6.6.5);
- 3. the premature start of the experiment command sequence on ground (Section 6.6.6).

Sadly, the systematic deviations of the experiment temperatures from their set point values went unnoticed in the pre-flight tests. Technically, it was a minor problem that might have been easily corrected if it had been recognised in time. Similarly, the overshoot of the centrifuge speed was, as a failure analysis revealed, due to an incorrect setting of a parameter in the centrifuge-controlling software. This mistake had also been overlooked in pre-flight tests. The premature start of the experiment command sequence on ground was due to a human error.

In conclusion, the flaws displayed on Biobox's maiden mission were all, one way or another, attributable to improper tuning and handling of an otherwise fine instrument. Except for the inherent lack of cooling capacity (see 6.3.9), which was recognised long before flight, the Biobox facility as such proved to be technically sound.

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7. The KNA Exposure Facilities

7.1 Introduction

Like Bion-8 and -9, the Bion-10 capsule carried four pan-shaped containers on its outer surface to accommodate exposure experiments. (According to some sources, all eleven Bion capsules were furnished this way.) The facilities were called KNA, which is short for *kontéjner naúchnoj apparatúry*, meaning container for scientific instruments.

The KNA is a cylindrical container, split in two parts (Figure 7-1). Its lower section (or base) is fixed to the capsule by six screws. Fastened to the base by a spring-loaded hinge is a lid that is held open in flight. The lower section and the lid each carry a detachable aluminum plate, 22 cm in diameter and 2.5 mm thick, onto which experiment packages can be strapped. In orbit, the packages are freely exposed to the conditions of open space. At the end of the flight the lid is closed, hermetically sealing the container for the re-entry. The outer surface of the KNA is covered with an ablative heatshield to protect the experiments against re-entry heat. The KNA is not re-usable.

7.2 Experiment Accommodation

The four KNAs on Bion-10 hosted a collection of exposure packages of international origin. Included were 14 ESA-IBMP experiment packages ($2 \times$ SEEDS, $12 \times$ DOSICOS). Their distribution over the plates mounted in the KNAs is given in Table 7-1.

7.3 KNA Integration Sequence

The integration of the experiment packages onto the four sets of two plates took place on



21 December (eight days before launch) at the School in Moscow. Holes were drilled into the plates as required by the footprints of the individual packages. The packages were then strapped onto the plates by copper wire, which was routed through the holes (Figure 7-2). The following day the loaded plates were flown to Plesétsk where the empty KNAs were already waiting.

On 27 December (two days before launch) all eight plates were mounted into the KNAs in a side room of the MIK, after which the KNAs

Figure 7-1. The KNA exposure facility. The spring-loaded lid is held open in orbit. Prior to re-entry, the lid is allowed to close under the action of the coiled spring, visible between the upper and lower sections. The protrusions on the lid contain catches that latch onto the lugs on the outside of the base. Two concentric white O-rings effect a hermetic seal between the lid and the base. Experiment packages are carried on detachable support plates as shown in Figure 7-2.

Table 7-1.			
	Place	Plate	Joint ESA-IBMP packages
KNA-1	bottom	B10-1	SEEDS 1b DOSICOS (IBMP: plastic)
	lid	B10-2	DOSICOS 3 DOSICOS 6
KNA-2	bottom	B10-3	SEEDS 1a DOSICOS 2 DOSICOS F1 DOSICOS (IBMP; depth dose)
	lid	B10-4	-
KNA-3	bottom	B10-5	DOSICOS 1 DOSICOS F2 DOSICOS (IBMP; depth dose)
	lid	B10-6	-
KNA-4	bottom lid	B10-7 B10-8	DOSICOS (IBMP; plastic) DOSICOS 4 DOSICOS 7

Table 7-1. Distribution of the ESA-IBMP experiment packages over the eight KNA plates. The plates are depicted in Figure 7-2. The individual experiment packages of DOSICOS and SEEDS are described in Sections 9-4 and 9-9.

were screwed onto the capsule (Figure 7-3). Around 13:00 h the first pair (KNA-1 and -2) was installed, followed much later, around 24:00 h, by the second pair (KNA-3 and -4). The reason for their late integration was that KNA-3 and -4 were positioned, in part, *over* the big hatch of the spacecraft. This pair of KNAs could be fixed only after this hatch had been closed, which did not take place until late at night, after all internal components of the payload had been integrated with the capsule. The position of the individual KNAs is sketched in Fig. 7-4.

Table 7-2. Summary of the postflight operations timeline of the exposure experiments.

Table 7-2.

7.4 The Opening and Closing Mechanism

The KNAs were mounted onto the spacecraft in the flight configuration, i.e. with their spring-loaded lids held open by retaining cables (Figure 7-5). The lid remained open throughout the launch and orbital flight. During launch, the experiment packages were protected only by the nosecone fairing of the launcher. At the end of flight, the retaining cables were released, and the KNA lids snapped shut. Four latches (Figure 7-1) held the lid in the closed position.

7.5 Attitude During Flight

The attitude of the Bion spacecraft is not controlled; the spacecraft spins slowly in orbit. The four KNAs would have pointed alternately towards the Earth and into deep space, throughout the flight in orbit.

7.6 Post-flight Operations

In the early morning of 10 January (Table 7-2) the spacecraft touched down in a forest in an ambient temperature of -22°C. The helicopters of the recovery team arrived almost immediately at the landing spot. Already at 07:22 h, six minutes after landing, IBMP's Vladimir Tsetlin was sitting on top of the capsule, warming his hands on the residual warmth of the surfaces of the KNA containers (estimated temperature: 40-50°C). Figure 3-12 shows two KNAs after landing, still in position on top of a Bion capsule. The KNAs were quickly detached, at the very landing site, whereupon the capsule was transferred by helicopter to a place nearby, outside the forest, where a tent was erected to protect the satellite and the recovery team against the cold.

In the evening of 10 January, around 19:00 h (when ESA's Biobox and the mini-facilities were already on their way to Moscow) the four KNA containers were flown by helicopter from the landing site to the airport of Karagandá, and from there to Kustanaj in the cargo bay of an Antonov An-12. Overnight,

















Figure 7-2. The eight KNA experiment plates, bearing the experiment packages, at the School in Moscow, prior to flight. The thinner packages were accommodated in the lids of the KNAs (A, C, E and G) and the thicker packages were mounted in the bases (B, D, F and H). All red parts were protective covers, removed before flight.

Figure 7-3. Mounting of KNA-2 on the spacecraft in the MIK at Plesétsk, two days before flight. The technician stands in front of the opening of the small hatch. The KNA is fixed onto the capsule by six screws. Note the silver colour of KNA's surface finish before flight. The lid is kept open, before the attachment of the retaining cable, by two pieces of white plastic. The screw holes prepared for KNA-3 and -4 can be seen in Figure 6-9.




Figure 7-4. Plan view showing the position of the four KNAs on the Bion-10 capsule. KNA-1 and -2 were placed above the small hatch and KNA-3 and -4 above the big hatch.

the KNAs were stored in a hotel in Kustanaj under the surveillance of Vladimir Tsetlin. The following morning, at 08:00, the KNAs were loaded into an Antonov An-24 aircraft. After waiting for some hours at the airport of Kustanaj, the aircraft took off for Moscow, landing at Chkalovskaya at 18:00 h. On Monday 11 January at 21:00 h (one and a half days after landing) the KNA containers were delivered at the School at 40, Shchukinskaya Street, the residence of the Radiation Research Institutes RCSRS and RSSS.

Source: Interview with V. Tsetlin by R. Demets 12 January 1993.

7.7 Post-Flight Inspection

The post-flight inspection of the KNAs and their experiments took place two days after landing at the School in Moscow (Figure 7-7). The following observations were made:

KNA-1

The lid was closed. When the release valve (a screw in the KNA bottom) was opened, air rushed into the KNA making a hissing sound, indicating that the KNA interior was still evacuated, as planned. All experiment packages were intact (Figure 7-8).



Figure 7-5. KNA-1 (right) and KNA-2 (left) in place on top of the re-entry capsule. The springloaded lid of each KNA is held open by a retaining cable which can be seen running down from the top of the lid.

KNA-2

The container was not completely closed; there was a gap visible between the lower section and the lid. Some experiment packages were scorched, in particular DOS 2, whose top was heavily burnt (Figure 7-9).

KNA-3

The container was not completely closed; there was a narrow gap visible between the lower section and the lid. DOS 1 was scorched and exhibited brown stains on its surface and small craters in the foil (Figure 7-10).

KNA-4

The container was closed, but not hermetically sealed. When the release valve was opened, no air was sucked in. The packages mounted in the lid of DOS 4 and DOS 7 were heavily scorched (Figure 7-11).

In summary, signs of damage to the experiment packages were detected in three of the four KNAs (Table 7.3).





Figure 7-6. The closing mechanism of a KNA. The spring-loaded lid is held open by a retaining cable that runs under a belt and is held in place by a knot, indicated by the arrow. Just before re-entry, the belt is released to liberate the re-entry capsule, thus freeing the cable and allowing the lid of the KNA to snap shut. The picture shows KNA-2, and the retaining cable of KNA-1 (see Figure 7-5). The position of the belts is illustrated in Figure 7-4.

Figure 7-7. Post-flight inspection of the four KNAs at the School in Moscow, 12 January 1993. From left to right: Yuri Akatov (RSSS), Lyudmila Nevzgodina (RSSS), René Demets (Hernandez Engineering), Aleksandr Vorozhtsov (RCSRS), Albert Marenny (RCSRS), Viktor Dudkin (RCSRS) and Vladimir Tsetlin (RSSS). Figure 7-8 (left). KNA-1 after flight. The foil wrap on the lid packages DOS-3 and DOS-6 was wrinkled and warped. Apart from that, the experiments, including SEEDS 1b, were recovered in good condition.

Figure 7-9 (right). KNA-2 after flight. The surface of DOS 2 was heavily scorched. DOS-F1 appeared to be intact. A dark streak was found on the goldcoloured hood of SEEDS 1a.







lid **†**

bottom





7.8 Temperature History

KNA-1 and KNA-4 were equipped with a temperature recorder provided by NASA. The recording range of these temperature loggers spanned from -40° C to $+60^{\circ}$ C. After the

Table 7-3.				
	Closed	Vacuum	Experiment status	
KNA-1	yes	yes	nominal	
KNA-2	no	no	damaged	
KNA-3	no	no	damaged	
KNA-4	yes	no	damaged	



temperature data had been downloaded at the School, two prints were provided to ESA, representing the temperature histories of the KNA-1 bottom and the KNA-4 lid. The plots show that temperatures swung wildly up and down over an amplitude that must have exceeded the recording limits (Figures 7-12 and 7-13). It is worth noting that in both plots the highest temperatures were recorded in orbit and not during the re-entry. In the base of KNA-1 (Figure 7-12) the temperature dropped below -40°C on 5 January (eight days into the flight). Peak values up to 60°C were recorded on 7 January (ten days into the flight). The highest temperature recorded during the reentry and landing phases was not more than

Table 7-3. Summary of the postflight inspection of the exposure experiments on 12 January.





T bottom flight. The foil wrap of DOS-1 had turned brown and it had acquired small craters in its surface. DOS-F2 appeared to be intact.

Figure 7-10 (left). KNA-3 after

Figure 7-11 (right). KNA-4 after flight. The surfaces of the two packages in the lid, DOS-4 and DOS-7 were scorched. In addition, the foil wrap of DOS-4 was ruptured. The detectors inside were mostly burned.

lid 🕇

↓ bottom

DS







10°C. (The ambient temperature at the landing site was -22°C).

In the lid of KNA-4 (Figure 7-13) a different history was recorded. First, the temperature never went below zero. Second, the temperature frequently reached 60°C or more. Third, the temperature was more unstable than that of KNA-1. The hotter history in the lid of KNA-4 may be explained by a stronger exposure to sunlight. The open lid may have received sunlight from a wide range of angles. In contrast, the bottom is shielded from above by the battery pack against solar rays, and from below by the capsule. The instability of

the lid temperature, as displayed in the plot, is indicative of a high degree of thermal decoupling, which allowed changes in incident solar radiation to be transformed immediately in temperature variations.

Particularly during the first week of the flight, the temperatures in KNA-1 and KNA-4 tended to move in opposite directions (when the one went up, the other went down and vice versa), which could have been due the fact that KNA-1 and KNA-4 were placed on opposite sides of the spacecraft (see Figure 7-4). When one side was in sunlight, the other was in shadow.



Figure 7-12. Temperature history in the lower part of KNA-1, as recorded by NASA.



Figure 7-13. Temperature in the lid of KNA-4 as recorded by NASA.

During the second week, the temperatures were higher than in the first week, just like the temperatures recorded inside the capsule. These measurements are consistent with the conclusion stated in Chapter 4 that, after one week, the spacecraft flew out of eclipse and from then on, was continually exposed to the Sun. Once back at ESTEC, the MLI hoods (thermal blankets) of the two SEEDS packages were further inspected. On the hood of SEEDS 1a (from KNA-2) a prominent streak was found, indicating exposure to high temperatures. The discolouration was limited to the outmost layer of the MLI blanket. The highest temperature seen by this foil was estimated at around 100°C. Whereas the scorched experiment surfaces in KNA-2 and KNA-3 may be explained by the failure of the lids to close prior to re-entry (Table 7-3) and the consequential lack of protection against the re-entry heat, the damage sustained by KNA-4 is more puzzling. KNA-4 was closed (though not hermetically sealed) and the temperature seems not to have exceeded 29°C during re-entry. One may conjecture that the damage to KNA-4 was not inflicted during re-entry but in orbit where the transient temperature excursions may have reached values far beyond the recording limit of 60°C (Figure 7-13).

7.9 Conclusions

Only one of the four KNAs (KNA-1) was retrieved under nominal conditions. The scorched experiment surfaces as observed in KNA-2, -3 and -4 are thought to be due to hot air blowing into the KNAs during re-entry, although it is possible that some overheating already took place in orbit prior to re-entry.

Whatever the reason, the damage to the experiments was unacceptably high. Similar problems had been encountered during the Bion-8 and Bion-9 missions. After the Bion-10 mission, ESA decided to cease using the KNAs and to change over to the more advanced Biopan European exposure facility which had successfully completed its qualification flight on Foton-8 in October 1992, just before the flight of Bion-10. After four successful missions since 1992, ESA's Biopan appears to be a more dependable and more versatile instrument than the KNA. Having said this, it should not be overlooked that the concept of Biopan was inspired by the KNA, and without the KNA, there would undoubtedly have been no Biopan.

8. Telemetry

8.1 Telemetry Format

The in-orbit performance of Biobox was followed by ESA by means of spacecraft telemetry, which was routed via the Flight Control Centre (TsUP; see Section 3.1.4) to Moslab. The TsUP was a military establishment to which ESA and its contractors had no access and thus a data link from TsUP to Moslab was logically the next best approach (Figure 8-1).

The telemetry provided by Biobox contained sufficient housekeeping data to determine whether the incubator was operating as planned. In case of an anomaly, a telecommand could be uplinked to Biobox. The Biobox telemetry consisted of discrete packets of data, which gave a status report every hour throughout the mission. Each packet contained 14 fields (message spaces) and these are listed in Table 8-1.

The first field, the header, indicated the start of the packet. The following three fields (Nos. 2-4) contained a serial number which identified the individual packets; this number was increased by one each time a new packet was transmitted. The ten remaining fields (Nos. 5-14) contained Biobox housekeeping data. Although some experiment-related information was included in fields 7, 10 and 14, no scientific data as such was provided in the telemetry.

8.2 Telemetry processing at TsUP

Following receipt by a ground station, the telemetry was forwarded to the TsUP, where it was reworked into computer files of two different types. The first type, identified by the

telemetry downlink path



letters DS, contained raw telemetry, while second type, identified by the letters BX, contained telemetry processed using conversion data provided by ESA. The DS and BX files were loaded into an IBM-compatible PC at the TsUP, which was linked to ESA's Moslab via a modem and a landline. In addition to the DS and BX files, a third file Figure 8-1. Overview of the telemetry data flow for Biobox on Bion-10.

14	лс 0-1.	
Nr	Message	Range
1	Header	not applicable
2	Telemetry package number: hundreds	0 to 9
3	Telemetry package number: tens	0 to 9
4	Telemetry package number: ones	0 to 9
5	Last two telecommands received	0 to 7
6	Power source: EGSE, mains or aux	0, 1 or 2
7	Temperature in the incubator (T9)	10 - 41°C
8	Temperature outside the incubator	0 - 62°C
9	Outer Peltier safety temperature	0 - 100°C
10	Centrifuge velocity	0 - 119 rpm
11	Centrifuge status: locked/unlocked; power on/off	0, 1, 2 or 3
12	Peltier current: positive=heating, negative=cooling	-10A to + 10A
13	Peltier status: on/off	0 or 1
14	Experiment status (5 independent groups: active or inactive)	0 - 31

Table 8-1. The 14 fields in theBiobox telemetry format

Table 8 1

type was provided by the TsUP. The latter, called NEWS, was issued once per day and contained a forecast of the next day's downlink times (see Table 4-7).

8.3 File Transfer From TsUP to Moslab

To transmit the files from the TsUP to Moslab, the Moscow telephone network was used. The files were downloaded from the TsUP computer only when the latter was polled by Moslab. In addition to the computer link, Moslab was connected to the TsUP by means of a voice link over the normal telephone line. This connection enabled personnel at Moslab to discuss the telemetry with representatives from IBMP (Viktor Golov) and KB Photon (Vladimir Kuznetsov) at the TsUP. However, Moslab was never in direct contact with the flight controllers.

8.4 More Telemetry

Apart from the Biobox telemetry, additional telemetry was made available to ESA, namely the ambient temperature inside the spacecraft as monitored by a thermal sensor, T6. The T6data were transmitted by telephone (a voice link) from the TsUP to IBMP personnel in Building 4, next to Moslab. The temperature history onboard the spacecraft, as measured by the T6, was replicated in Building 4's programmable incubator which contained the ground reference experiments of ALGAE, CLOUD, DOSICOS, FLIES, SEEDS and WOLFFIA.

8.5 Coverage

Biobox's first telemetry packet (No. 001) was issued one day before launch, with Biobox integrated in the satellite which was then already on the launch pad. This was followed by another 22 pre-launch packets. During orbital flight, 276 packets (Nos. 024 through 299) were downlinked, with the final packet issued three hours before landing. Out of these 276 packets, 242 were relayed by TsUP to Moslab. ESA received the remaining 34 packages (12%) only after completion of the mission. Of the 242 telemetry packets provided to Moslab, 117 were converted into in the BX format, and the remaining 125 were sent as raw data.

8.6 Delay Time

The telemetry packets arrived in Moslab after a considerable delay. The delay time was quite variable, ranging from a couple of hours up to almost a full day. The three major factors contributing to the duration and variability of the delay time were the spacecraft downlink pattern, the telemetry processing by the TsUP and the inefficiency of the Moscow telephone network.

8.6.1 Spacecraft downlink pattern

Every hour the spacecraft computer commanded Biobox to produce a fresh telemetry packet; Biobox responded by delivering an update of the 14 fields in series, along a single serial analogue line. The new information was not immediately relayed to Earth, but was instead recorded on tape and stored onboard until a ground station commanded the spacecraft to downlink its data. The downlinking occurred predominantly between 09:00 h and 18:00 h Moscow time, in a series of six data dumps equally spaced over that period (Figure 8-2). The telemetry packets produced by Biobox after 18:00 h were stored on-board overnight and dumped during the first pass the following morning. In other words, after six p.m., the TsUP (and also Moslab) received no new Biobox telemetry over a period that usually lasted more than 15 hours (Figure 8-2).

8.6.2 File preparation by TsUP

Once the telemetry arrived at the TsUP, it was not immediately accessible to the ESA team. The new data were first screened by the TsUP personnel. Even when TsUP provided raw data only (the DS files) a substantial amount of time could pass before the data became available to the ESA team.

From 3 to 10 January, detailed records were kept at Moslab about the times that Biobox files were available for downloading from the TsUP computer. These times were then compared with the times at which the ground station lost contact with the spacecraft. This information was provided by the TsUP in the NEWS files. From this survey we learned that the time that passed between the downlink sessions to the ground stations, and the availability of the files in the TsUP computer, varied from 1.5 to 7.5 hours.

8.6.3 Performance of the Moscow telephone link

A third contributor to the delay time was the Moscow telephone network, which was used to link Moslab with the TsUP. During the seven-day period 3 to 9 January 1993, 111 attempts were made by Moslab to poll the TsUP computer, of which only 31 resulted in the computer answering the call. However, three of these attempts were immediately frustrated when the line was disconnected before the login procedure could be completed. Of the 28 successful logins, only 12 sessions ended with a normal logoff; in 16 cases an early exit resulted when the connection was interrupted by factors beyond the control of the team at Moslab. During six of the 16 premature terminations, the interruption occurred while a file transfer was in progress.



Once the data were flowing over the Moscow telephone line, the rate of transmission was quite good. Detailed records of the modem and software download speeds, measured in characters per second (CPS), were kept for the period 6 to 9 January 1993. Over that four-day period, a total of 39 files were downloaded with an average speed of 194 CPS.

8.7 Accuracy of the Telemetry Data

To determine the reliability and accuracy of the telemetry that ESA received in Moslab, the data supplied by the TsUP were compared after flight with the data stored in the Biobox RAM-card memory. The period of time selected for evaluation was the Biobox "RUN" mode, which started on 29 December when Biobox reached the experiment operating temperature and ended on 7 January just before the cooling of the incubator started. This period was chosen for two reasons. Firstly, with its duration of nine days, "RUN" covered a substantial and representative part of the mission and secondly, during this phase of the operations, the Biobox temperatures were quite stable. The incubator temperature, as monitored by the Biobox T9 sensor, was used for the analysis. This temperature was given in field 7 of the telemetry packet (Table 8-1).

Figure 8-2. ESA/MOSLAB was provided by the TsUP with a daily forecast of the next day's downlink sessions (Table 4.7). In total, date and time of 49 sessions were announced (red dots). On most of the mission days the spacecraft dumped its data in a regular pattern of six sessions, evenly spread between 09:00 and 18:00 Moscow local time. No forecast was provided for 29 December, nor for 8, 9 and 10 January. For 30 December, a string of eight downlink sessions was announced, but only five were time-tagged. Details of only one session were presented for 1 January. On three occasions, telecommands were uplinked to Biobox (TK1, TK2 and TK3, see Table 6.5). Note that the timing of the telecommands (white circles) fits the downlink pattern; obviously, spacecraft passages over ground stations were simultaneously used for up- and downlinks. The total time that Bion-10 spent in orbit is indicated by shading.

Table 8-2: Comparison between Biobox incubator temperature data as stored in the Biobox memory (Ram Card) and as provided by telemetry (DS files: raw data; BX files: data processed by TsUP). The number of samples "n" reflects the number of data points available from that particular data source.

Table 8-2.		
Data Sourc	e	Run Phase
		temperature (°C)
Ram Card		$37.54 \pm 0.11 \ (n = 211)$
Telemetry	DS files	$37.07 \pm 0.17 (n = 189)$
	BX files	$37.05 \pm 0.15 \ (n = 100)$

Table 8-2 contains a summary of the calculations performed on the available data. It appears that the incubator temperature as reported by the telemetry was systematically half a degree Celsius lower than that derived from the RAM-card data, which suggests that some data errors may have been introduced in the data link. In addition, the standard deviation was slightly higher in the spacecraft telemetry data compared to the results obtained with the RAM-card. It should be noted however that the RAM-card data were more precise anyway, having a resolution of 0.2°C versus 0.5°C in the telemetry. No detectable loss or distortion of information seemed to occur during the processing of data from DS files into BX files.

8.8 Conclusions

With 88% of the telemetry packages transmitted to Moslab, the telemetry coverage was adequate, or even good. The same positive conclusion can be drawn about the accuracy of the data. The weak point was definitely the tardy transfer of telemetry to Moslab. The purpose of the computer file transmissions to Moslab was to provide ESA with an "early warning system" for recognising anomalies in the in-flight performance of Biobox which would facilitate decision making, in conjuction with the TsUP, in respect of the telecommands to be sent to Biobox. The telemetry received in Moslab in the form of computer files never fulfilled this objective, firstly because the information was simply too old by the time it arrived, and secondly because anomalies reported in the Biobox telemetry had been spotted and analysed by the flight controllers at the TsUP before ESA had even seen the data.

On the positive side, through the computer link with the TsUP, Moslab was provided with direct print-outs of the innumerable data that Biobox produced during flight. In terms of reliability, this was a vast improvement over the simple voice link transfer used for Bion-8 and Bion-9.

Main source:

Telemetry analysis: Biobox on Bion-10 Arthur Knell, ESTEC (GPS), 5 April 1993

9. The Experiments

9.1 ALGAE

Title:

Changes in the cell division cycle of Chlamydomonas monoica caused by microgravity **Experiment identifier:** BIOK-88-6-NL **Study object:** Unicellular green algae (species: Chlamydomonas monoica) **Investigators:** H. van den Ende, Universiteit van Amsterdam (NL) E. van Spronsen, Universiteit van Amsterdam (NL) M.L. van den Briel. Universiteit van Amsterdam (NL) O.V. Gavrilova, University of St. Petersburg (RUS) Funding: Netherlands national funding (SRON)

9.1.1 Summary

The ALGAE experiment was an investigation into the division cycle of the unicellular green alga Chlamydomonas monoica under conditions of weightlessness. Twenty-four liquid cultures of C. monoica, each 600 µl, were grown for four days under a diurnal regime of 14 h light and 10 h darkness. At the end of the culturing period, the cells were chemically fixed. After flight, the number and size of the cells were compared with reference samples taken from a control experiment that had been conducted on-ground under ambient gravity conditions (1g). The experiment was severely affected by the abnormal temperature rise that occurred within the spacecraft during the latter part of the mission and, in particular, by the ensuing premature return of the spacecraft to Earth. When the capsule landed, the experiment was still in progress, with none of the samples fixed. As a result, no conclusive data could be gained about the influence of microgravity on the alga's cell cycle.

9.1.2 Scientific objectives

The cell cycle of C. monoica can be synchronised by growing the cells under alternating conditions of light and dark. In a perfectly synchronised culture, all cells are at the same stage of their cycle at any point in time. Originally entitled Effects of microgravity on plasma membranecytoskeleton interactions during cell division of C. reinhardtii, with the emphasis on ultrastructural details in dividing cells, the focus of the ALGAE experiment shifted towards basic cell cycle parameters like cell number and cell size as the experiment approached its readiness for flight. In the process, C. reinhardtii was replaced by C. monoica for the practical reason that zygospores of the latter species are more easily obtained in the laboratory.

Preliminary indications that *Chlamydomonas* might proliferate faster in weightlessness came from an experiment by O.V. Gavrilova and A.V. Gabova flown on Bion-9 in 1989 [1].

9.1.3 Biological materials

Chlamydomonas monoica is a unicellular, homothallic, green alga. Upon nitrogen starvation – accomplished in the laboratory by presenting the cells with a culture medium devoid of nitrogen salts – the alga produces germ cells (gametes) which after sexual fusion (fertilisation) develop into zygospores. Zygospores are thick-walled zygotes, capable Figure 9-1. The ALGAE facility with the cover off. Size: $106 \times 124 \times 175$ mm. The cell cultures were contained in four mix units (the upright blocks with printed circuit boards on the outside). Photo: CCM. of maintaining their viability for a prolonged period of time under adverse environmental conditions. Zygospores can be induced to germinate by providing nutrients plus light, upon which the vegetative growth cycle is resumed.

The ALGAE flight experiment contained 24 cell cultures, each consisting of 20000 to 25000 zygospores suspended in 600 µl of distilled water.

9.1.4 Chemicals

Culture medium The algae were grown in Bold's basal medium of the following composition:

NaNO ₃	0.250 g/l
NaCl	0.025 g/l
MgSO ₄ .7H ₂ O	0.075 g/l
$CaCl_2.2H_2O$	0.025 g/l
K ₂ HPO ₄	0.095 g/l
KH ₂ PO ₄ .3H ₂ O	0.175 g/l
NaHCO ₃	0.4 mM
trace elements (TES D)	1 ml/l
final pH	6.6

. The Fixative

Figure 9-2. The mix unit. The ALGAE facility carried four of them. Illustration: CCM.

Samples were fixed in 2% (final concentration) paraformaldehyde.





9.1.5 Experiment hardware

The ALGAE facility (Figure 9-1) weighed 3.5 kg. It was developed and manufactured by the Dutch company CCM as a fully automated culturing device which, after being primed before launch, was able to complete an intricate experiment protocol in a completely autonomous way. It was equipped with four battery-powered, electronically operated, cell culturing modules (known as "mix units") each of which contained six cell cultures of 600 µl (Figure 9-2). Each cell culture was connected with two 600 µl reservoirs, one containing culture medium, the other a fixing solution. The transfer of these fluids to the cultures was pre-programmed and effected by spring-loaded pistons that were released on receipt of an electrical signal. The 24 cultures were individually illuminated by red light (660 nm wavelength), produced by lightemitting diodes. These provided the energy that allowed the photosynthetic algae to grow. The synchronisation of the cell cycle was accomplished by applying a diurnal regime of 14 h light and 10 h darkness, causing the algae to alternately increase in size when exposed to the light and divide when in the dark.

The temperature was monitored by three internal thermometers. As the ALGAE facility was not equipped with a thermal control system, the cell cultures were passively exposed to the ambient temperature in the Bion capsule. In fact, the temperature of ALGAE increased to slightly above ambient because of the heat produced by its lightemitting diodes. The experiment history, which included records of the deployment of the pistons, the temperature history and switching on and off of the light-emitting diodes, was stored in an electronic memory whose contents were downloaded after flight.

9.1.6 Mission timeline events

The mix units were filled with zygospores at the University of Amsterdam one week before flight and then flown to Moscow at L-5 days. Integration of the units with the ALGAE facilities (flight and ground) was done by CCM at Moslab at L-4 days. The electrical power of ALGAE was switched on just before departure for Plesétsk, at L-3 days. Transport to Plesétsk was in a thermally controlled box maintained at 22°C.

The experiment was primed to be active during the last part of the spaceflight, with the objective of reducing the on-board storage period of the fixed cultures to a minimum. (The quality of the fixed cells decreases with time, especially if not stored in a refrigerator.)

During the first nine days of flight the spores were kept in darkness and deprived of nutrients to keep them dormant, whereafter a four-day period of proliferation was initiated by the supply of illumination and of nutrients. The cultures were subjected to four cycles comprising alternate exposures to 14 h of light and 10 h of darkness. Individual cultures were scheduled to be fixed in darkness at 2, 4, 6 and 8 h after the final 14 h period of light, when the cell number was expected to have reached its plateau.

After landing, ALGAE was removed from the capsule at 10:25 (L + 3 h 9 min). To prevent the fixed cultures from decaying during transport back to Moslab, the facility was packed in a thermal box at the landing site and maintained at a temperature of 4° C until it reached its destination.

9.1.7 Results

Technical performance of the ALGAE facility The experiment history was fully recorded in the electronic memory. Stored data indicated that all fluid transfers and illumination cycles had been executed according to plan. Four of the 24 cultures were prevented from growing when fixative spread from a single leak.

Impact of the thermal anomaly and early return of the spacecraft

The temperature rise in the Bion-10 spacecraft and its premature return to Earth interfered with the ALGAE experiment in several ways:

- ALGAE was active during the last days of the flight, when the temperature in the capsule went above 30°C. This temperature was far beyond the range 22-24°C, specified for optimal growth of *C. monoica;*
- as planned, the ALGAE facility was stored in a cool box after landing. However, with the flight cut short by two days, the experiment was still in progress at that point of time. The algae were returned to normal gravity conditions, and cooled down to 4°C, before being fixed;
- the pre-programmed transfer of formaldehyde to the cultures was performed after landing with the experiment in the cool box at 4°C. The cold increased the viscosity of the fixative, impeding its smooth transfer to the cultures and affecting the quality of the fixations.

Scientific results

The post-flight investigation revealed an unusually high degree of variability. Germination (estimated by the number of zygospores, i.e. the number of non-germinators) had successfully taken place in some cultures, but was disappointing in others. Similarly, the rate of cell proliferation, as judged by the total number of cells retrieved from the individual cultures, was highly inconsistent. On top of this variability, a number of technical flaws were identified:

- leakage of fixative from one location had impaired the growth of four cultures;
- in some cultures, bacterial infections were detected;
- the fixations appeared to have suffered from the temperature drop (see also above).

In conclusion, only a few of the 24 cultures could be regarded as having undergone a successful germination *and* a successful proliferation *and* a successful fixation.

Back in Amsterdam, the ultrastructure of the cell bodies was investigated, including the pyrenoid, a protein body embedded in the chloroplast. The pyrenoid is associated with starch synthesis and is surrounded by deposits of starch; it is one of the principal reserve food materials in green algae as well as in green plants. The examination revealed evidence that the average size of the pyrenoid was substantially larger in the space-flown cultures compared to the ground controls.

In a slightly modified form, the ALGAE experiment was reflown on Foton-10 (1995), Foton-11 (1997) and Foton-12 (1999).

9.1.8 Scientific co-operation

The post-flight analysis was a collaboration between the University of Amsterdam (The Netherlands) and the University of St. Petersburg (Russia). The two universities had complementary responsibilities, Amsterdam being responsible for the CSLM analysis and St. Petersburg for the TEM analysis. Within the framework of the ALGAE experiment, Olga Gavrilova (St. Petersburg), whose pioneering work led to the flight of *Chlamydomonas* on Bion 9 [1], worked for some time in Amsterdam.

9.1.9 Publications

The results of the ALGAE experiment on Bion-10 have not been published externally.

Table 9.1. BONES sample configurationexperiment unitsbonesflight microgravityn = 7n = 56flight 1g (centrifuge)n = 2n = 16ground 1gn = 7n = 56totaln = 16n = 128Each plunger unit carried 2 x 4 = 8 foetal long-bones.

9.2 BONES

Title:

Growth and mineralisation in isolated foetal mouse bones under microgravity conditions **Experiment identifier:** IMMU-88-8-NL Study object: foetal mouse long bones **Investigators:** J.P. Veldhuijzen, ACTA, Vrije Universiteit, Amsterdam (NL) J.J.W.A. van Loon, ACTA, Vrije Universiteit, Amsterdam (NL) B. Zandieh Doulabi, ACTA, Vrije Universiteit, Amsterdam (NL) C.M. Semeins, ACTA, Vrije Universiteit, Amsterdam (NL) N.V. Rodionova, SIZ*, Ukrainian Academy of Sciences, Kiev (UA) O.P. Berezovska, SIZ*, Ukrainian Academy of Sciences, Kiev (UA) Funding:

Netherlands national funding (SRON)

* I.I. Shmalgausen Institute of Zoology

9.2.1 Summary

In the BONES experiment, the influence of weightlessness on bone development was investigated. Embryonic long bones, dissected from mouse foetuses, were cultured *in vitro* over a period of four days. The matrix mineralisation appeared to be suppressed in space, particularly in the samples exposed to microgravity, whereas the growth of bone length was unaffected.

9.2.2 Scientific objectives

Weakening of the skeleton of astronauts during spaceflight due to demineralisation is a frequently reported, but poorly understood phenomenon. The loss of bone mass is generally thought to be due to a direct reaction of the bone tissue to the "unloading" of the skeleton under conditions of weightlessness, but other spaceflight-related factors like psychological stress, the re-distribution of body fluids and changes in the hormone

Table 9-1. Sample configuration

of BONES. Each experiment

unit carried $2 \times 4 = 8$ foetal

mouse long bones.

balance may contribute as well. To eliminate such indirect factors, the BONES experiment was conducted on *in vitro* cultures of mouse bones. Surgically dissected, and kept alive in a culture medium, such bones are literally cut off from any external signals produced by the body.

9.2.3 Biological materials

The bones used were the mid-foot long-bones called metatarsals. These are rod-like bones belonging to the hind-foot of tetrapod vertebrates, with usually one corresponding to each toe. The metatarsals used for the experiment were dissected from mouse embryos that had been isolated from pregnant mothers (strain: "Swiss Random") at 17 days of gestation. At this age, the bones are about 1.5 mm long and mainly composed of cartilage. Through mineralisation, i.e. the deposition of calcium phosphate into the cartilage matrix, the metatarsals finally develop into mature bones, a process that can be partly followed in vitro. A total of 128 metatarsals was used in the BONES experiment, of which 72 were launched into space (Table 9-1).

9.2.4 Chemicals and materials

Culture medium

The bones were cultured in Alpha MEM medium, without nucleosides, which contained the following supplements:

NaHCO ₂	7.2 g/l
gentamicin	50 ug/l
fungizone	0.5% v/v
Na-β-glycerophosphate	3.0 mM
vitamin C	50 ug/l
L-glutamin	300 ug/l
BSA Factor-V	2 9/1
final pH	7.3

Atmosphere

The ambient atmosphere was air containing 5% carbon dioxide.

Fixative

Samples were fixed in 0.5% (final concentration) paraformaldehyde.

9.2.5 Experiment hardware

BONES was one of the four experiments that

flew in Biobox. As was the case for the other Biobox experiments (OBLAST, MARROW and FIBRO) the hardware consisted of the units described in Section 6.3.1. The main material used for the construction of the units of the BONES experiment was PET (polyethylene tetraphtalate), an opaque, white plastic. Each plunger unit was equipped with two 1-ml culture compartments, in each of which four bones were grown. The culture compartments were covered by a gaspermeable foil made of polyethylene. The BONES units were packed in sealed experiment containers filled with air containing 5% carbon dioxide.

9.2.6 Mission timeline events

The BONES experiment was prepared in Moslab, where a mouse vivarium had been set up. Two weeks before launch, the first batch of mice was brought from Amsterdam to Moscow. One week later, a second batch arrived. Five days before launch, the metatarsals were dissected from the embryos. Four days before launch, the metatarsals were loaded into the plunger units. The loaded plunger units were maintained in Biobox at 20°C until launch to prevent the bones from growing before exposure to weightlessness.

The active phase of BONES spanned the first four days of the flight. After one hour in orbit the bones were supplied with fresh culture medium. After two days, the medium was once more replenished. After four days of culturing, the bones were chemically fixed.

9.2.7 Results

Technical performance

All 96 fluid transfers (six per experiment unit) were executed as planned. After flight, a significant amount of air was found trapped in several culture compartments although no evidence for leakage was found.

Impact of the thermal anomaly and early return of the spacecraft

The thermal anomaly in Biobox (see Section 6.6) occurred when the active phase of the experiment was terminated, with all bones chemically fixed. The uncontrolled storage temperature at the end of the flight had no

Table 9-2. Summary of the results of BONES.

		Increase in Length (%)	
	number of bones	overall	diaphysis
ground 1g	n = 9	21.1 ± 2.6	143.9 ± 17.3
flight 1g (centrifuge)	n = 12	11.5 ± 1.7	93.4 ± 4.5
flight (microgravity)	n = 6	17.1 ± 2.5	76.7 ± 5.2

serious impact on the quality of the fixed bones, compared to the nominal scenario.

Table 9-2. BONES: Summary of results.

Scientific results

To quantify their development, two parameters were measured for each individual bone, before and after the flight:

- the overall length;
- the length of the diaphysis (the mineralised centre of the bone).

The post-flight values (Lpost) were compared with the pre-flight values (Lpre). The increase in length, ΔL , was expressed as a percentage according to the following formula:

 $\Delta L = \frac{Lpost - Lpre}{Lpre} \times 100 \%$

Unfortunately, the bones were recovered in a poor condition. The main reason must have been the lengthy pre-launch storage period at 20°C in Biobox. With a duration of more than three days, this was at the very limit of what the bones could endure. A significant amount



of necrotic tissue was displayed and it appeared that the majority of the 128 bones had not increased in length whatsoever. These bones had to be excluded from the evaluation. Not more than 27 bones passed this selection. These had grown by between 10% to 25% during the four-day culture period, but no convincing differences were revealed when the three groups of samples (ground, flight and flight-centrifuge) were compared. In contrast, the diaphysis seemed to have undergone a prominent increase in size, with substantial differences discernible between the three groups The strongest increase was observed at 1g on ground, the weakest in microgravity, with the in-flight centrifuge group assuming an intermediate value (Table 9-2).

The following conclusions could be drawn:

- the overall growth in microgravity was not demonstrably different from the overall growth at 1g;
- 2. the mineralisation of the diaphysis was reduced by the spaceflight, the more so when the bones had been exposed to microgravity.

The credibility of the Bion-10 results was considerably enhanced by the fact that the BONES team observed very similar trends – reduced mineralisation with unchanged length growth – in two Biorack experiments on the US Space Shuttle in 1992 [2, 3] and 1994 [4]. Owing to superior late access conditions, in these Biorack experiments the bones were recovered in a much healthier state (Figure 9-3).

9.2.8 Scientific co-operation

Out of every eight bones in an experiment unit, one bone was reserved for electron microscope analysis, to be performed in Kiev by

Figure 9-3. The growth of the mineralised centre of the foetal bone (the diaphysis) was reduced in microgravity. This was consistently observed by the BONES team on three consecutive missions (Biorack on IML-1 in 1992, Biobox on Bion-10 in 1992/93, and Biorack on IML-2 in 1994). N.V. Rodionova and her co-workers. Indications were found that the bones' chondrocytes might proliferate and differentiate more slowly in microgravity. However, owing to the small number of usable samples, it was not possible to draw firm conclusions about the effects of microgravity on the bone ultrastructure and mineral composition.

9.2.9 Publications

The experiment gave rise to the following publications:

Berezovska O.P., Karmozina L.G., Rodionova N.V., Veldhuijzen J.P. 1994. *Is calcification of fetal bone responsive to microgravity?* Cospar '94 Book of Abstracts p 286, 30th Cospar Scientific Assembly, Hamburg, Germany.

Rodionova N.V., Veldhuijzen J.P., 1994. Razvítie embrionál'noj kósti in vitro v uslóviyakh kosmícheskogo polëta (Development of foetal bone *in vitro* during spaceflight [in Russian] *Space Biology and Aerospace Medicine*, Abstracts of the Tenth Conference, Moscow, p 102.

Van Loon J.J.W.A., Berezovska O., Zandieh Doulabi B., Semeins C.M., Rodionova N.V., Veldhuijzen J.P., 1995. *Reduced mineralisation in isolated fetal mouse long bones flown on board the Russian Bion-10 satellite*. In: Effect of spaceflight and hypergravity on mineral metabolism in organ cultures of fetal mouse long bones, PhD thesis, J.J.W.A. van Loon, Vrije Universiteit, Amsterdam.

Veldhuijzen J.P., Bervoets T.J.M., Blaauboer C.W., Diedonné S.C., Haaijman A., Hagen J.W., Semeins C.M., Zandieh Doulabi B., 1996. *Isolated skeletal tissues cultured under microgravity*. Proceedings of the Cosparmeeting, p 307. Birmingham, UK.

Veldhuijzen J.P., van Loon J.J.W.A., Bervoets T.J.M., Blaauboer C.W., J.P. Duke, Haaijman A., Montufar-Solis D., Semeins C.M., Zandieh Doulabi B., Growth and mineralization in cultured fetal mouse long bones under continuous and daily interrupted microgravity. Results of two spaceflight experiments (to be published).

9.3 CLOUD

Title:

The impact of pre-flight gravity stress on in-flight fitness in *Drosophila melanogaster*

Experiment identifier:

Study object:

Fruit flies (species: *Drosophila melanogaster*) Investigators:

I.A. Ushakov, Laboratory of Gravitational Biology, IBMP, Moscow (RUS)
A.M. Alpatov, Laboratory of Gravitational Biology, IBMP, Moscow (RUS)
N. Tarakanova, Laboratory of Gravitational Biology, IBMP, Moscow (RUS)
R. Marco, Universidad Autónoma de Madrid (E)
Funding:

IBMP (RUS)

9.3.1 Summary

Three populations of fruit flies, pre-adapted for more than ten generations to different gravity environments (1g, 10g and 1gclinorotation at 8 rpm) were tested for their capability to produce offspring during spaceflight. Each group was found capable of doing so. A correlation between pre-flight gravity regime and in-flight performance could not be assessed.

9.3.2 Scientific objectives

The experiment CLOUD (the name being derived from a cloud, or swarm, of flies) was an attempt to modify the in-flight fitness of fruit flies by acclimatising the insects pre-flight to altered gravity conditions.

A few months before flight, a genetically heterogeneous population of wild-type fruit flies was divided into three parts. The first group, the control, was maintained at 1*g*; the second group was kept at 10*g* on a low-speed centrifuge and the third group was subjected to rotation at 8 rpm on a low-speed clinostat.

Table 9-3. Fruit fly life cycledata.

In a clinostat, the test object is rotated about the horizontal axis such that the magnitude of the gravity vector is maintained at an average value close to 1g, while the vector's direction, with respect to the test object, is continually changing. Biological objects on a clinostat sometimes behave as if exposed to weightlessness.

The exposure to these three different g-environments was maintained for more than ten generations. The assumption was that, in the process, the three groups would start to diverge genetically by natural selection (survival of the fittest), eventually producing three ecotypes with a specific fitness towards respectively 1g, 10g and clinorotation at 1g. (An ecotype is a group within a species adapted genetically to a particular habitat but able to cross freely with other ecotypes of the same species.)

The expected result was that, in space, the clinostat-adapted group would perform best, and the 10*g*-adapted group worst.

9.3.3 Biological materials

Three groups of fruit flies (species: Drosophila melanogaster, strain: wild-type Oregon-R) were used, each one acclimatised to a different gravity environment. Each group was represented in flight by duplicate sets of 20 females and 20 males (number of flies per group: $2 \times 40 = 80$, total number of flies: $3 \times 80 = 240$). As shown in Table 9-3, D. melanogaster Oregon-R develops from embryo into adult in ten days, which is, within the nominal duration of the Bion flight.

9.3.4 Culture medium

The fruit flies were raised on an agarsolidified culture medium. Propionic acid was added as a preservative.

agar	2.0%
cane sugar	3.6%
wheat flour	3.6%
yeast extract	10.7%
propionic acid	1.1%

9.3.5 Experiment hardware

The CLOUD device was developed and manufactured to the specifications of

Table 9-3. Fruit fly life cycle data

life cycle stage	duration
embryo (egg)	1 day
larva	5 days
pupa	4 days
imago (adult)	60 days (median)

Dr. Ushakov. The manufacturer was a company in St. Petersburg, called Biofizpribór, a specialist in hardware for life science experiments in space. The result was a robust, 5.1 kg instrument, largely made of steel, fitted with six drum-shaped modules in which the fruit flies could move, eat, develop and reproduce.

A remarkable feature was that the fruit flies' progeny, emerging in space, could be segregated in flight from their parents, so that the generations could separately be analysed after flight. This was accomplished by installing feeder trays (used by the fruit flies not only as dinner table, but also as egg depot) that could mechanically be rotated; during rotation, the eggs were moved out of the parent's chamber into another chamber. Each of the six fruit fly modules was equipped with three such chambers and two feeder trays. The rotations were executed by an electromotor which received its power (27 V DC) from the spacecraft batteries. The motor was geared in such a way that the feeder trays would move synchronously in all six modules. The insects were exposed to alternate 12-hour periods of light and darkness by a set of 12 light bulbs, to prevent possible microgravityinduced aberrations in their circadian system. The tray movements and the light-darkness cycles were pre-programmed in, and commanded by, an external electronics unit. A single thermometer was installed with a link to the spacecraft's telemetry system, allowing the in-flight temperature to be monitored on the ground. CLOUD was encased in a rectangular metal box measuring $270 \times 218 \times 102$ mm (1 × $w \times h$). This box was mechanically attached to the satellite structure by four bolts (Figure 9-4).

It is worth noting that, years later, a concept comparable to that of Ushakov's, which allowed the segregation of successive generations, was adopted for a device called "Multigeneration Cultivation for Insects", now under development for use in ESA's Biolab onboard the International Space Station.

9.3.6 Mission timeline events

The three groups of fruit flies were prepared at IBMP Plánernaya in Building 4. Three days before launch, the fruit flies were transferred from Moscow to Plesétsk. Installation of the flies into the CLOUD device took place two days before launch, in "the Hut", IBMP's quarters in Myrnij (Figure 9-5).

The following sequence of events reflects the *planned* experiment scenario (identical for each of the six modules); it was envisaged that after flight, chamber 1 would be occupied by first generation individuals, chamber 2 by the second generation and chamber 3 by the third.

First feeder tray shift

20-30 minutes after launch, feedtray 1 would be presented to chamber 1. The flies would deposit their eggs in the feedtray. The tray would remain for 12 hours in this position, collecting eggs.

Second feeder tray shift

Filled with eggs, feedtray 1 would be transferred to the still-vacant chamber 2. During the following 10 days, the eggs would develop into adult flies (the second generation).

Third feeder tray shift

On the last day of the flight (scheduled for L + 13d) feedtray 2 would be presented to chamber 2, while feedtray 1 was withdrawn. The second generation would have laid its





eggs in feedtray 2. The feedtray would remain for 12 hours in this position, collecting eggs.

Fourth feeder tray shift

Just before landing (scheduled to take place at L + 13.5d) feedtray 2 would be shifted to chamber 3 so that the eggs produced by the second generation would be segregated. These eggs would develop into third-generation flies. This third generation would not have progressed beyond the egg and larva stage by the end of the flight.

The CLOUD ground control experiment was conducted at IBMP Plánernaya in Building 4, in a duplicate model of the CLOUD facility, with a 24-hour delay with respect to the flight experiment.

9.3.7 Results

Technical performance of the CLOUD facility Owing to a technical failure, the planned experiment scenario did not proceed beyond the first feeder tray shift. (Note that even if this failure had not occurred, the third and fourth shifts would not have taken place in space in view of the premature landing of the spacecraft). Figure 9-4. Configuration of the CLOUD facility.

Figure 9-5. Two days before launch, CLOUD underwent its final preparations in "the Hut", a wooden villa in Myrnij (Plesétsk). Table 9-4. The first generationafter flight. The flies werecounted at the landing site,immediately after landing.Pre-flight number of adults permodule: 40.

Table 9-4.	
Pre-flight treatment	Surviving adults per module
10g centrifuge	n = 30 n = 27
clinostat (8 rpm)	n = 18 n = 23
1 <i>g</i>	n = 5 n = 8

Impact of the thermal anomaly and early return of the spacecraft

The temperature rise in the Bion-10 capsule and its premature return to Earth disturbed the CLOUD experiment scenario in the following ways:

- during the second half of the flight the temperature rose to levels that were detrimental to the health of the fruit flies;
- with the flight cut short by two days, the CLOUD experiment was still in progress when the capsule landed.

Scientific results

Within three and a half hours after touchdown, the CLOUD device was removed from the Bion capsule and opened by Dr. Ushakov at the landing site. It was only then that the malfunction was detected. In all six modules only the first feeder tray shift had been completed, so that the planned segregation of generations had not been accomplished. From each module adults were recovered, accompanied by embryos, larvae and pupae, demonstrating that flies from each of the three groups had been able to produce offspring in space. From the fact that black, ready-to-hatch pupae were not observed in any module, it was

Table 9-5.	
Pre-flight treatment	Number of eggs per female
10g centrifuge clinostat (8 rpm) 1g	13.8 ± 0.75 12.6 ± 2.5 16.7 ± 4.2

concluded that the imagines belonged to the first generation, and the embryos, larvae and pupae to the second.

To quantify the in-flight fitness of the flies, two parameters were investigated:

- the number of survivors after flight,
- their fecundity.

The survival rate of the first generation appeared to be the highest in the 10g group and the lowest in the 1g group (Table 9-4).

To assess the fecundity of the three groups, all second-generation individuals were counted, namely embryos, larvae and pupae. From this number, the average number of eggs laid by the first-generation females could be deduced. It seemed that the 1g group had performed best in space, although the statistical significance of the data was low (Table 9-5).

Since the feedtray movements in the control experiment conducted on ground had been executed according to the *nominal* scenario, flight and ground data could not readily be compared. In fact, numerous additional ground control experiments were performed before, during and after the mission. Despite all of these efforts, none of these yielded results that could prove, or disprove, that the three groups of flies had indeed acquired a specific fitness for the gravity environment to which they had been exposed before flight. Nor was it possible to detect a consistent difference between the results obtained in space and on-ground.

9.3.8 Scientific cooperation

The initial concept was to distribute the samples equally between the IBMP- and ESAaffiliated investigators. (Note that the CLOUD device contained two identical sets of three modules). Eventually, a different scheme was adopted whereby the complete experiment was analysed in Spain, with Ilya Ushakov temporarily joining the team of Roberto Marco in Madrid.

9.3.9 Publications

The results of the CLOUD experiment have not been published externally.

Table 9-5. Eggs laid in space per1st-generation female.

9.4 DOSICOS

Title:

Identification and quantification of incident radiation particles during orbital spaceflight **Experiment identifier:** BIOK-88-9-D (second reflight)

Study object:

Space radiation

Investigators: G. Reitz, DLR, Cologne (D) R. Facius, DLR, Cologne (D) M. Schäfer, DLR, Cologne (D) R. Beaujean, University of Kiel (D) W. Heinrich, University of Siegen (D) A.I. Vikhrov, RCSRS, Moscow (RUS) V.E. Dudkin, RCSRS, Moscow (RUS) A.M. Marenny, RCSRS, Moscow (RUS) A.B. Akopova, RCSRS, Moscow (RUS) Yu.V. Potapov, RCSRS, Moscow (RUS) Yu.A. Akatov, RSSS, Moscow (RUS) V.V. Arkhangelsky, RSSS, Moscow (RUS) Funding:

Funding:

Russian (IBMP) and German (DLR) national funding

9.4.1 Summary

DOSICOS was a multi-facetted radiation dosimetry experiment. By using detectors of various types, different segments of the space radiation spectrum were monitored. All detectors but one were placed on the outer surface of the spacecraft, where they would be exposed to the space environment where shielding was minimal. Although some valuable data were obtained, the scientific return of DOSICOS was significantly reduced owing to detector damage caused by overheating.

9.4.2 Scientific objectives

The experiment DOSICOS (short for "Dosimetry on Cosmos") was a combination of four dosimetrical studies:

- measurements of the spectral composition of the heavy ion (HZE) component of the radiation field;
- measurements of the total radiation dose outside and inside the capsule;
- neutron measurements (secondary radiation);

 depth-dose measurements: determination of the penetration depth of different particles, and the decrease of the dose in dependence of mass shielding.
 The DOSICOS experiment flew earlier in 1987 (Bion-8) and 1989 (Bion-9). The repetition at 2-3 years intervals would allow

the investigators to monitor temporal variations in the radiation environment, particularly in relation to the state of the eleven-year solar cycle. The flight of Bion-8 took place near a solar minimum, Bion-9 and Bion-10 were close to a solar maximum.

9.4.3 Detectors

A thirteen-piece set of dosimeter packages was used; this consisted of five "universal" stacks, four neutron detectors and four depth-dose stacks (Figure 9-6). The total experiment mass was about 550 g. Twelve detector packages, wrapped in Kapton foil, were accommodated in KNA containers where they would be directly exposed to the space environment (see Chapter 7). A single stack, encased in a Type I container, was installed inside the spacecraft on the floor alongside the WOLFFIA experiment.

Universal dosimeters (5x)

A combination of three detector types was used to measure the flux of cosmic particles accumulated over the entire flight as well as their characteristics (charge=Z, energy=E, linear energy transfer=LET. The first comprised an arrangement of plastic sheets of different composition and registration thresholds for tracking and detecting heavy ions (LET range: from 100 MeV/cm to 10 GeV/cm). The second type were nuclear emulsions for detecting all incident particles coming directly from space, (i.e primaries including protons) as well as secondary particles (neutrons, nuclear disintegration events). The third type were thermoluminescence detectors (TLD) for measuring the total absorbed dose. Three packages were provided by ESA and another two by IBMP (Figure 9-6).

Neutron dosimeters (4x)

The dosimeters used for neutron measurements consisted of TLDs of different isotopic composition (TLD 600 and TLD 700 crystals) embedded in polyethylene foil,



Figure 9-6. The DOSICOS experiment consisted of thirteen radiation detector packages of different types. All but one (DOS 5) were outside-exposed under minimal shielding conditions. complemented by four plastic detector sheets, each of a different material (CR39, CNK, CND and Lexan (Figure 9-6).

Depth dosimeters (4x)

Two depth-dose stacks were prepared for ESA by DLR, another two by RCSRS and RSSS for IBMP. The ESA stacks were composed of 11 slices (TLD 700), those of IBMP consisted of 16 slices (TLD 600).

9.4.4 Materials

Materials used in the detectors were as follows:

TLDs

lithium fluoride; Emulsions silver halide crystals embedded in gelatine (trade name: K2 and K5,

manufacturer: *Ilford*, *UK*);

Plastic foils

diallyl diglycol carbonate (trade name: CR39); cellulose nitrate (trade name: CNK, manufacturer: Kodak, France); cellulose nitrate (trade name: CND, manufacturer: Daicel, Osaka, Japan); polycarbonate (manufacturer: General Electric, USA, trade name: Lexan); *Envelope*

polyimide plastic film (trade name: Kapton H, manufacturer: DuPont, USA).

The composition of the materials used in the Russian detectors is not known in detail. Some alternative plastic foil materials including CN85, Lavsan and CR29 seem to have been used, as well as a Russian-made cellulose nitrate referred to as TsN. The nuclear emulsions were designated as BR-2 and BYa. The thermoluminescence detectors in the Russian depth dose stacks were made of lithium fluoride (TLD 600).

9.4.5 Mission timeline events

The total exposure time was 11.6 days (11 d, 14 h, 22 min). The exposure of the external detectors started when the nosecone fairing was released on 29 December at 16:33 h. The KNA containers were closed at the moment that the re-entry capsule was released from the service module on 10 January at 06:55 h.

9.4.6 Results

Impact of the thermal anomaly and early return of the spacecraft

Judging by the scorched and burnt appearance of their surfaces after flight, many DOSICOS packages must have seen some extremely high temperatures during the mission (see Chapter 7). There are indications that the high temperatures were experienced in orbit, before re-entry. During the second half of the flight, when the spacecraft flew out of eclipse, the temperature in the lid of KNA-4 frequently went beyond the +60°C limit of the temperature recorder (see Chapter 7).

The early return of the spacecraft meant that DOSICOS was exposed to space radiation for 11.6 days instead of 13.6 days, but this reduction did not essentially affect the experiment.

Thermal damage during re-entry

The DOSICOS packages were distributed over all four KNA containers. Three of the four KNAs were not completely closed during re-entry. As a consequence, many DOSICOS packages were not properly protected against the re-entry heat (see Chapter 7).

Owing to the damage caused by overheating, only part of the DOSICOS detectors were suitable for analysis after flight.

Depth-dose measurements

A good set of depth dose data was obtained, both from ESA and from IBMP (Figure 9-7). In ESA's depth dose stack F2 the shielding ranged from 51 mg/cm² up to 2.1 g/cm², spread over 11 steps. This was achieved using three thin thermoluminescence detectors (98 mg/cm²) plus eight normal detectors (240 mg/cm²). The midpoint value of the top detector including its cover of 13 µm kapton foil was 49 mg/cm² + 2 mg/cm² = 51 mg/cm². The maximum recorded dose rate was 3 Gy per day (behind 51 mg/cm²) falling off to 0.25 mGy per day (behind 2.1 g/cm²) (Figure 9-7).

In IBMP's depth dose experiment a similar shielding range was covered in 16 steps. In this case, Kapton foil (4 mg/cm²), eight thin detectors (~ 60 mg/cm²) and eight normal detectors (260 mg/cm²) were used. The maximum dose rate was 2 Gy per day, falling off to 0.25 mGy per day (Figure 9-7).

The Bion-10 mission took place close to a solar maximum. When the measured depth dose values were compared with theoretical models of the space radiation environment AP-8 (protons) and AE-8 (electrons), a good fit was found with the conditions pertaining at solar maximum. These models predicted the dose rate as function of shielding thickness,



taking into account the contributions by the radiation belt and by background radiation from space.

Total dose measurements

Inside the capsule, total dose values ranging from 2.31 up to 3.05 mGy were measured, corresponding to 199-263 μ Gy per day (Table 9-6).

Linear energy transfer measurements

From measurements taken by IBMP, an average linear energy transfer of the HZE particles of 12.4 ± 2.7 keV/µm in tissue was determined. This value would cause a radiation quality factor of 2.5 to 3.5 based on ICRP Report No. 26 (published by the International Commission for Radiological Protection, Stockholm).

Under the names MAPPING and DOSIMAP, follow-on experiments of DOSICOS were performed under comparable conditions in orbit by G. Reitz *et al.* in ESA's Biopan that flew on Foton in 1994, 1997 and 1999.

9.4.7 Scientific cooperation

The ESA- and IBMP-affiliated investigators

Figure 9-7. Results of the DOSICOS depth-dose measurements on Bion-10. Over the first 2.5 g/cm² in front of the detector, the dose rate steeply declined by four orders of magnitude.

Table 9-6.

	<u>TLD 700</u>		<u>TLD 600</u>	
Sheet	total dose	dose rate	total dose	dose rate
	(mGy)	(µGy/d)	(mGy)	(µGy/d)
DOS-5 (top)	2.54 ± 0.25	219	2.96 ± 0.11	255
DOS-5 (bottom)	2.97 ± 0.35	256	3.05 ± 0.06	263
WOL FELA	2.31 ± 0.08	199	2.87 ± 0.03	247
WOLITIA	2.51 ± 0.08	199	2.87 ± 0.05	247

Table 9-6. Total dose and (calculated) daily dose values as measured in the DOS 5 stack. Additional TLDs were placed in the WOLFFIA stack, see Section 9-10. Both stacks were placed in the same container, labelled SEEDS/ DOSICOS/WOLFFIA (see Figure 2-7). worked closely together during most phases of the experiment, i.e. the design, preparation and calibration of the detectors, data exchange and discussion of the spectra obtained by use of different theoretical models. Tasks were distributed as follows:

ESA

General coordination:	G. Reitz
Nuclear emulsions:	R. Facius, M. Schäfer
TLDs:	R. Facius, M. Schäfer
CN and Lexan analysis:	R. Beaujean
CR39 analysis:	W. Heinrich

IBMP

General coordination:	A.I. Vikhrov
HZE measurements:	A.M. Marenny
Neutron dosimetry:	Yu.V. Potapov
Depth dosimetry:	Yu.A. Akatov
Other tasks:	A.B. Akopova
	V.V. Arkhangelsky
	V.E. Dudkin
	V.I. Popov
	A.S. Vorozhtsov

9.4.8 Publications

The results of the depth-dose experiments can be found in the following publications:

ESA:

Depth Dose Distribution Measurements, G. Reitz, R. Facius, P. Bilski, P. Olko (2002) (in press).

IBMP:

Absorbed Dose Measurements and LET Determination with TLDs in Space, Vana N., Schöner W., Fugger M., Akatov, Y. (1996) *Rad. Protection Dosimetry* **66**, Nos. 1-4, pp. 145-152.

9.5 FIBRO

Title:

Comparative study of morphophysiological properties and differentiation of cell culture fibroblasts under conditions of spaceflight (microgravity) and on Earth **Experiment identifier**:

Study object:

Foetal mouse fibroblasts

Investigators:

M.G. Tairbekov, IBMP, Moscow (RUS) A.V. Gabova, IBMP, Moscow (RUS) L.B. Margolis, M.V. Lomonosov State University, Moscow (RUS) B.A. Bajbakov, M.V. Lomonosov State University, Moscow (RUS) Funding:

ESA

9.5.1 Summary

Cultures of mouse fibroblasts were flown in space to investigate the influence of microgravity on the morphology, locomotion and proliferation of these cells. The cells acquired a more rotund shape, their nuclei became significantly smaller and rounder, while the direction of locomotion became more irregular. The rate of cell proliferation remained unchanged.

9.5.2 Scientific objectives

Cells of connective tissue (fibroblasts) were cultured under microgravity conditions to see if this would have an effect on their morphology and development. The cultures came in two varieties. The first type, called histoculture, consisted of integral pieces of connective tissue dissected from a mouse embryo. In these cultures, the fibroblasts were maintained in their natural three-dimensional micro-environment of extracellular matrix materials. The second type, called monolayer, was a two-dimensional culture in which a single layer of isolated fibroblasts was spread out over a glass surface. It was thought that the two culture types might behave dissimilarly under changes in gravity conditions because in the histocultures, gravity was expected to exert a tension on the extracellular matrix that might stimulate the fibroblasts, whereas in the monolayers, the fibroblasts were deprived of this putative external sensory network.

9.5.3 Biological materials

Histocultures

Small blocks of connective tissue, typically a cubic millimetre in volume, were obtained from mouse embryos (strain: A-Sn) on the seventh day of gestation. These blocks were then anchored onto a sheet of gelfoam.

Monolayers

The monolayer fibroblasts were obtained from a mouse embryo (strain: F-208) at the eighth day of gestation. The cells were cultured for one passage in a Petri dish, and then spread out on glass cover slips measuring 22×9.5 mm by 0.2 mm thick. The cells were scraped away from both ends of the coverslip to create two free zones (see Figure 9-8). Monolayered fibroblasts are known to respond to such "wounding" by migrating into the evacuated zone, and filling the gap.

9.5.4 Chemicals and materials

Culture medium

The fibroblasts were cultured in RPMI 1640 medium (Sigma R 7006) to which 10% serum had been added.

Radioactive marker for DNA replication During flight, the cultures were presented with fresh medium to which ³H thymidine was added at a concentration of 4 μ Ci per ml.

Buffer

The cells were rinsed in Hank's balanced salts solution (HBSS; Sigma H 6136) before being fixed.

Fixative

Glutaraldehyde (final concentration: 3.0% v/v)

Cell supports

Histocultures: gelfoam (sponge); monolayers: glass coverslips.

9.5.5 Experiment hardware

FIBRO was one of the four experiments that

flew in Biobox. As was the case for the other Biobox experiments (BONES, MARROW and OBLAST), the hardware consisted of the units described in Section 6.3.1. The main material used to construct the FIBRO units was PSU (polysulphone), a semi-transparent, yellowbrown plastic. Each FIBRO plunger unit was equipped with two 1-ml culture compartments, without provision for gas exchange. The FIBRO flight set consisted of three units. A replica set of three was kept on the ground as a 1*g* reference. Each FIBRO experiment unit hosted two cultures (Table 9-7).

9.5.6 Mission timeline events

Ten days before launch, the fibroblasts were isolated. Four days before launch, the FIBRO cultures were loaded into the experiment units at the Moscow State University. The same day, the units were transported to Moslab for integration into Biobox. Prior to launch, the temperature of the fibroblasts was kept at 20°C to suppress growth and development. In orbit, their temperature was raised to 36.5°C. The experiment protocols of the monolayers and the histocultures were different, as detailed below:

Monolayers

Two hours after launch the monolayers were presented with fresh medium containing tritiated thymidine. 48 hours later, the cultures





Table 9-7.				
		Histocultures	Monolayers	Location in
				Biobox
Flight (μg)	experiment unit A	2	_	CIS-2
	experiment unit B	_	2	CIS-1
	experiment unit C	1	1	CIS-3
Ground $(1g)$	experiment unit D	2	_	CIS-2
	experiment unit E	-	2	CIS-1
	experiment unit F	1	1	CIS-3

Table 9-7. Sample configurationof FIBRO.

Table 9-8. Size and shape of the

cell nuclei in the FIBRO

cultures.

were rinsed in buffer and, 10 minutes afterwards, fixed in glutaraldehyde.

Histocultures

One-and-a-half days after launch, the histocultures were presented with fresh medium containing tritiated thymidine. Three days later the cultures were rinsed in buffer and, 30 minutes thereafter, fixed in glutaraldehyde.

9.5.7 Results

Technical performance of the FIBRO experiment units

All FIBRO experiment units performed nominally; all 36 fluid transfers (six per plunger unit) were executed as planned. After flight, a leak was detected in experiment unit B, with about 0.5 ml of fluid spilled.

Effect of the thermal anomaly and early return of the spacecraft

The thermal anomaly in Biobox (see Section 6.6) occurred after all FIBRO cultures had been fixed. The uncontrolled storage

Table 9-8.		
Monolayers	µg (n=409)	1g (n=400)
Length x 10 ⁻² mm	0.224 ± 0.002	0.333 ± 0.004
Width x 10 ⁻² mm	0.167 ± 0.002	0.231 ± 0.003
Histocultures	µg (n=808)	1g (n=800)
Length x 10 ⁻² mm	0.129 ± 0.001	0.154 ± 0.002
Width x 10 ⁻² mm	0.099 ± 0.001	0.116 ± 0.011

temperature in Biobox did not significantly affect the fixed cultures, which, in any case, had been planned to experience high temperatures after fixation. The early return of the capsule Earth had no effect on FIBRO.

Cell density

After flight, the cell density of the monolayers exposed to microgravity was substantially lower than those of the ground controls. Although this seemed to point at a reduced rate of proliferation in space, this hypothesis was not corroborated by the number of ³H-labeled nuclei, an indicator of the fraction of dividing cells. This fraction appeared to be the same in the monolayers exposed to microgravity and to 1g (Figure 9-9). Other explanations are possible. Cells may have become detached by the launch vibrations, or the cell-to-surface adhesiveness may have detoriated in microgravity, resulting in cells breaking loose from the substrate.

Shape of the cell body

The shape of the cells in the histocultures exposed to microgravity was not markedly changed. By contrast, the fibroblasts in monolayers exposed to microgravity had adopted a less elongated, more rotund and contracted morphology, compared to the ground controls.

Shape and size of the nucleus

In the monolayer cultures exposed to microgravity, the nuclei were significantly smaller (surface area reduced by about 50%) and at the same time more rotund. Similar tendencies were observed in the histocultures (Table 9-8).

Cell migration in the monolayer cultures Whereas the 1*g* controls displayed straight wound edges with cells migrating straight into the free zones (as judged by their shape, with their longitudinal axis perpendicular to the cutting edge), the space-flown cultures showed edges that were more blurred, with cells apparently moving more randomly into the free zone.



9.5.8 Scientific cooperation

Although FIBRO was a proprietary IBMP investigation, the experiment was firmly rooted in the joint ESA-IBMP programme as exemplified by the fact that it flew on ESA's Biobox in ESA-provided experiment units. Furthermore, ESA-affiliated investigators joined IBMP's team for FIBRO follow-on flights on Foton-10 (1995) and Foton-11 (1997).

9.5.9 Publications

Gabova A.V., Tairbekov M.G., Margolis L.B., Bajbakov B.A., 1994. Cell culture in microgravity conditions (Experiment FIBROBLAST), Cospar '94 Book of Abstracts p 286, 30th Cospar Scientific Assembly, Hamburg, Germany (1994).

Tairbekov M.G., Margolis L.B., Bajbakov B.A., Gabova A.V., Dergacheva G.V., 1994. Rost i podvízhnosť klétok v kuľtúre (in vitro) v uslóviyakh mikrogravitatsii (Eksperiment "Fibroblást"), growth and motility of cell culture in microgravity conditions (Experiment Fibroblast), [in Russian]) *Izv. Russian Acad. Sci.* (ser. biol.) 5, pp 745-750.

Tairbekov M.G., Gabova A.V., Margolis L.B. Bajbakov B.A., 1994. Vliyánie mikrogravitátsii na rost i podvízhnosť klétok fibroblástov v kuľtúre (in vitro) (Microgravity effects on the growth and mobility of fibroblast cells in culture (in vitro) [in Russian]), Abstracts of the Xth Conference Space Biology and Aerospace Medicine, p 87, Moscow.

Tairbekov M.G., 1997. Issledovaniya na kul'túre klétok shivótnikh in vitro (eksperiment "Fibroblast") in: Gravitatsiónnay Biológiya Klétki (teóriya i eksperimént), pp 82-91, ISBN 5-207-00-455-14.

9.6 FLIES

Title:

Test of the metabolic hypothesis of accelerated ageing in Drosophila in space ESA experiment identifier BIOK-88-14-E, BIOK-88-15-E **Study object:** Fruit flies (Drosophila melanogaster) **Investigators:** J. Miquel, Universidad de Alicante (E) E. de Juan, Universidad de Alicante (E) R. Marco, Universidad Autónoma de Madrid (E) I.A. Ushakov, Laboratory of Gravitational Biology, IBMP, Moscow (RUS) A.M. Alpatov, Laboratory of Gravitational Biology, IBMP, Moscow (RUS) Funding:

Spanish National Space Programme ("Accion Especial")

9.6.1 Summary

FLIES was an attempt to verify, in a single experiment, two earlier independent findings regarding young male fruit flies: enhanced motor activity during spaceflight and a shortened life span after landing. Both phenomena were successfully reproduced. A novel feature of FLIES was the automated in-flight monitoring of the locomotor activity of the insects.

9.6.2 Scientific objectives

In a NASA experiment flown previously on Bion-3, evidence was found that the life span of young male fruit flies was shortened after flight, while mature flies (two-week old at launch) were not much affected [5]. Videorecordings from a later ESA experiment on the US Space Shuttle suggested that young fruit flies turn exceptionally active in space: an excited, accelerated type of motility was observed [6]. To combine these findings a synthesising hypothesis was formulated: in space, the sexual activity of young male fruit flies is enhanced; the concomitant higher rate of oxygen consumption makes the insect grow old at a higher speed (cf. the "free radical" ageing theory [7]). In the FLIES experiment

Figure 9-9. Number of nuclei in the FIBRO monolayer cultures per field of view under the microscope at a magnification of 7×90 . flown on Bion-10, this theory was put to the test by measuring, in a single experiment, the in-flight activity of fruit flies in conjunction with the post-flight lifespan. Two populations were compared: "adults" (eclosed on Earth) versus "youngsters" (eclosed in space). To test the alleged link between enhanced activity and sexual behaviour, male-only populations were compared with mixed populations comprising males and females.

9.6.3 Biological materials

Fruit flies (species: *Drosophila melanogaster*) of the Oregon-R strain were used, at two different stages of development:

- youngsters: pupae, 3-4 days old at the time of launch;
- adults: imagos, 11 days old at the time of launch.

Typical duration of the stages in the life cycle of a fruit fly:

embryo (egg) 1 day; larva 5 days; pupa 4 days; imago 60 days (median).

9.6.4 Experiment hardware

The FLIES experiment was conducted in a fully automated miniaturised facility, weighing 2.5 kg which was manufactured by the Ondatron company of Madrid according to the requirements of the investigators. Called the DEMIR-108 (DEMIR = DEtection of Motility by InfraRed), it was tailored to fit inside a



Russian BB container (Figure 9-10). The DEMIR-108 was able to monitor and record the locomotor activity of eight groups of flies over a three-week period. It contained the following subsystems:

Culture tubes

The fruit flies were housed in 16 polystyrene culture tubes (8 cm long and 2.5 cm in diameter), closed by cotton plugs (Figure 9-10). Each tube contained 30 flies on agar-solidified feeding medium. The 16 tubes were divided in two sets of eight, with each set consisting of:

- 2 tubes with 15 males and 15 females (imagos);
- 2 tubes with 30 males (imagos);
- 2 tubes with 15 males and 15 females (pupae);
- 2 tubes with 30 males (pupae).

In one set, the motor activity of the flies was continually recorded before, during and after flight. The flies from the other set were removed from the DEMIR after landing and subjected to behavioural tests.

Monitoring equipment

In the first set, each culture tube was equipped with a single light-emitting diode that emitted a beam of infrared light (invisible for fruit flies) across the tube, aimed at a sensor at the opposite side of the tube. The motility of the flies was quantified by counting the interruptions of the light beam.

Batteries

The light-emitting diodes and the light sensors were powered by a 16-piece array of 3-volt lithium cells that provided enough energy to keep the monitoring equipment alive for at least 21 days (Figure 9-10).

Recording equipment

The recording equipment consisted of four commercial ACR SR-009 pulse loggers (one logger per two tubes). Every five minutes, the accumulated number of light beam interruptions over that period was read, timetagged and stored in the logger's memory. Each logger was provided with two internal temperature sensors whose outputs were recorded along with the activity of the flies.

Figure 9-10. The DEMIR-108, with the cover off. Size: 106 x 124 x 175 mm. It contained 16 culture tubes for the fruit flies (the picture shows one of these tubes). The activity of the fruit flies was monitored by infrared light. The light-emitting diodes and light sensors (not shown in the photograph) were powered by 16 lithium batteries, located in the upper part (the red blocks). Data were recorded by four loggers located in the compartment at right. The loggers were powered by their own internal batteries.

Mechanical structure

The tubes, the monitoring equipment, the battery pack and the data recorders were mounted in a simple aluminum frame (Figure 9-10).

9.6.5 Culture medium

The 5.5 cm-long culture tubes were filled to a depth of 2 cm with a feeding medium (Figure 9-10) which consisted of:

- 12% castor sucrose;
- 0.6% propionic acid (preservative);
- 100 µg/ml streptomycin (antibiotic);
- 100 µg/ml penicillin (antibiotic);
- 2-4% agar (solidifier).

The medium was devoid of proteins and amino acids to prevent the females from laying eggs. This was because larvae, emerging from the eggs during flight, tend to turn the culture medium soft while feeding, giving rise to imagos being trapped in the medium and being killed. See also reference [8].

9.6.6 Mission timeline events

The fruit flies were bred in the laboratory at Alicante. Six days before launch, the flies were transported to Moscow. The DEMIR was loaded at IBMP in Building 4 at four days before launch. Transport to Plesétsk took place three days before launch in a thermal box at 10°C (the low temperature was selected to suppress the development of the pupae before flight). The DEMIR's monitoring equipment was switched on two days before launch in Plesétsk.

The motor activity of the flies was recorded non-stop from two days before launch to one day after landing. The experiment was returned to IBMP within 17 hours of landing, upon which the recorded data were immediately downloaded.

The ground reference experiment was conducted in a duplicate DEMIR at IBMP in Building 4, but delayed 24 h with respect to the flight experiment.

9.6.7 Results

Technical performance DEMIR-108 performed well without any problem.

Impact of the thermal anomaly and early return of the spacecraft

The FLIES experiment was not significantly invalidated by the temperature rise onboard the spacecraft, nor by the early return of the capsule to Earth.

Scientific results

In each of the four groups flown in space, a significant motor activity was displayed, whereby the activity profile of the young flies was exceptionally high and much more pronounced than that of the adult flies. As this difference was true for both the male-only series and the mixed series, there was no indication of a direct link with mating behaviour. In fact, as of today, the exact reason for the hyperactivity of young male fruit flies in space remains unknown.

The expected accelerated ageing after spaceflight was confirmed by two separate test results: the mating capability declined at a higher rate, and the flies died faster than usual. The results from the FLIES experiment were in agreement with the metabolic hypothesis of accelerated ageing in *Drosophila* in space, though by themselves insufficient to prove the causal relation ("high activity makes life shorter") postulated by this theory.

In a slightly modified form, the FLIES experiment was reflown on Foton-10 (1995) and Foton-11 (1997), confirming and extending the results from Bion-10.

9.6.8 Scientific cooperation

The FLIES experiment was jointly prepared by investigators from ESA and IBMP, with the hardware provided by the Spanish team. The postflight analysis took place in Madrid, with Ilya Ushakov temporarily joining the Spanish team.

9.6.9 Publications

The results from the FLIES experiment on Bion-10 have not been published externally.

9.7 MARROW

Title:

Effect of microgravity on in vitro cultures of pre-osteoblast-like cells ESA experiment identifier BIOK-88-16B **Study object:** MN7 (osteogenic cell line) **Investigators:** G. Schoeters, VITO, Mol (B) J. Bierkens, VITO, Mol (B) J. Maes, VITO, Mol (B) N.V. Rodionova, SIZ*, Ukrainian Academy of Sciences, Kiev (UA) O.P. Berezovska, SIZ*, Ukrainian Academy of Sciences, Kiev (UA) Funding: Belgian national funding (PRODEX)

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9.7.1 Summary

Cultures of osteoblastic cells were grown for nine days under weightlessness. The osteogenic activity of the cells, as judged by the production of alkaline phosphatase and collagen Type I, was found to be suppressed. In contrast, if the cells were presented with IL-1 (interleukin-1) or PTH (parathyroid hormone), the synthesis of alkaline phosphatase and collagen Type I was higher in microgravity that under 1*g*. The rate of cell proliferation was not affected.

9.7.2 Scientific objectives

The phenomenon of bone mass reduction in space is thought to originate at the cellular level, where the bone mass budget is controlled by the antagonistic efforts of osteoblasts (bone-forming cells) and osteoclasts (bone-degrading cells). The MARROW experiment aimed to investigate whether the proliferation rate and the boneforming capabilities of osteoblasts are influenced by weightlessness.

The MARROW experiment used *in vitro* cell cultures to eliminate the many external factors interfering with cell proliferation and

differentiation *in situ*. The culturing period was nine days, sufficiently long to permit (at least at 1g) the cells to differentiate into mature bone producers. Part of the cultures was stimulated with IL-1 (interleukin-1) or PTH (parathyroid hormone) to see if, under weightlessness, the responsiveness of the cells to these effector molecules was changed. After flight, cell culture extracts were examined for ALP (alkaline phosphatase) and collagen Type I as markers for osteogenic activity.

9.7.3 Biological materials

The original MARROW experiment proposal was written around work previously performed by Schoeters et al. [9] on in vitro mineralisation of marrow plugs isolated from mouse femurs. Preflight tests however revealed that these plugs were easily destroyed by the launch vibrations. As a replacement, a murine cell line named MN7 was adopted [10], whose osteoblastic features include the ability to produce ALP, collagen Type I and osteocalcin. The cells were grown as threedimensional cultures on collagen sponges in aliquots of 3 x 10^6 cells per sponge. When grown on collagen, the pre-osteoblast-like MN7 cells are induced to differentiate, whereby the capabilities to produce ALP, collagen Type I and osteocalcin are sequentially acquired. There were 36 MARROW cultures in flight and 28 reference cultures on ground (Table 9-9).

9.7.4 Experiment hardware

MARROW was one of the four experiments that flew in Biobox. As was the case for the other Biobox experiments (BONES, OBLAST and FIBRO), the hardware consisted of the units described in Section 6.3.1. The main material used for construction of the units comprising MARROW was transparant polycarbonate. Each plunger unit was equipped with two 1-ml culture compartments, each holding two cell cultures on collagen supports. The MARROW plunger units were accommodated in sealed experiment containers with an internal atmosphere consisting of 5%carbon dioxide in air. The culture compartments were covered by a gas-permeable foil made of polyethylene, permitting the exchange of carbon dioxide with the environment.

9.7.5 Chemicals and materials

Culture medium

The cells were cultured in serum-free BGJb medium (Gibco 041-2581) with the following supplements:

- 1% gentamicin (antibiotic);
- 2 mM L-glutamine;
- 10^{-2} M ß-glycerophosphate.

The final pH value was 7.4.

Radioactive tracer

Collagen synthesis was measured by the incorporation of ³H proline into collagenasedigestible protein. There was 3μ Ci tritiated proline per ml culture medium.

Growth factors

IL-1 (interleukin-1) at 1 ng/ml PTH (parathyroid hormone) at 10⁻⁷ M

Cell supports

Collagen Type I (Porous Collagen Matrices M1531)

Atmosphere

The ambient atmosphere was air containing 5% carbon dioxide.

Fixative

PLP (paraformaldehyde/lysine/periodate) at 0.1%.

9.7.6 Mission timeline events

Culturing of the MN7 cells began in the laboratory of VITO at Mol (B), two weeks before launch. One week before launch, the cultures were flown to Moscow. The loading of the experiment units took place in Moslab and was completed at three and a half days before launch. In Biobox, the units were maintained at 20°C until launch.

Table 9-9.		
	Experiment	Cell
	units	cultures
Flight microgravity	n= 7	n= 28
Flight 1g (centrifuge)	n= 2	n= 8
Ground 1g	n= 7	n= 28
Total	n= 16	n= 64

The active phase of MARROW spanned the first nine days of the flight. All MARROW cultures were simultaneously provided with fresh medium at day 4, with fresh medium plus labelled proline and stimulants at day 7, and with fixative at day 9.

9.7.7 Results

Technical performance

MARROW was the only Biobox experiment that suffered from failing plunger activations, albeit at a modest degree: 94 out of 96 fluid transfers were executed as planned. The two failures occurred in one unit from the microgravity flight series.

Impact of the thermal anomaly and early return of the spacecraft

The thermal anomaly in Biobox (described in Section 6.6) happened when the active phase of the experiment was terminated, with all cultures chemically fixed. As a late marker for osteoblastic differentiation, the production of osteocalcin was planned to be measured. The uncontrolled storage conditions after day 9 caused the osteocalcin degrade to un-detectable levels despite the early return of the capsule.

Scientific results

Cell proliferation was measured by the protein and DNA contents at day 9. No significant differences were found between the 1g and microgravity series. Cell differention was judged by the expression of collagen Type I and ALP at day 9. In microgravity, the production of both compounds was significantly suppressed (Figure 9-11). However, cells presented with interleukin-1 or PTH appeared to have produced much more collagen Type I and ALP than their counterparts that had been exposed to 1g(Figure 9-12). In short, the osteogenic activity in the cultures exposed to microgravity was unusually low in the absence, and unusually high in the presence, of stimulating agents.

After the completion of the Bion-10 mission, the MARROW project was passed on from VITO to the University of Leuven (B). Under the aegis of the new investigator team,

Table 9-9. Sample configuration of MARROW. Each plunger unit carried $2 \times 2 = 4$ cultures.



Figure 9-11. Effect of 9 days of microgravity on the production of ALP and collagen in 15-day old MN7 cultures. Ground: ground control experiment; 1g centrifuge: inflight control experiment; flight: samples exposed to microgravity. Source: Schoeters *et al.* (1994).

Figure 9-12. Effect of IL-1 (1ng/ml) and PTH (10⁻⁷ M) on the production of ALP and collagen in MN7 cultures. Cultures were stimulated for 2 days, 7 days after microgravity was reached. Flight: samples exposed to microgravity; ground: ground control experiment; centrifuge: inflight control experiment. Source: Schoeters *et al.* (1994). follow-on MARROW experiments were flown on Biobox-2 (1995), Biobox-3 (1997) and Biobox-4 (1998). In these experiments the original MN7 cells were replaced by MG63 cells.

9.7.8 Scientific co-operation

After flight, four cultures (two non-stimulated flight cultures and two non-stimulated ground cultures) were handed over to Natalia Rodionova and co-workers for ultrastructural analysis with a transmission electron microscope in Kiev (UA). These cultures were fixed in 2.0% glutaraldehyde in cocadylate buffer at pH 7.4. The Kiev team found indications of an underdeveloped Golgi apparatus and endoplasmatic reticulum in microgravity, in agreement with the retarded osteogenic activity observed by the Mol team. The limited amount of sample material however prevented statistically sound conclusions from being drawn.

9.7.9 Publications

Bierkens J., Maes J., Ooms D., Collier M., Vangenechten C., Borremans B., Van Vlasselaer P., Schoeters G., 1993. *Decreased acquisition of osteoblastic phenotype markers and increased response to interleukin-1 and parathyroid hormone in pre-osteoblast like cells under conditions of microgravity. In:* Proceedings of the 5th European symposium on life sciences research in space, Arcachon, France, 26 September - 1 October 1993. ESA SP-366, pp 25-30. Noordwijk, The Netherlands: ESA Publications Division.

Schoeters G., Bierkens J., 1994. *Effects of Microgravity on Bone Cell Differentiation*. In: Space Scientific Research in Belgium Vol 1 Microgravity, pp 89-94.

Berezovskaya O.P., Karmozina L.P., 1994. Ul'trastruktúrnyj análiz osteogénnykh klétok MN7, kul'tivírovannykh b nevesómosti. (Ultrastructural analysis of osteogenic MN7 cells cultured in microgravity). (In Russian). Abstracts of the Xth Conference Space Biology and Aerospace Medicine, Moscow, 1994 p 87.



9.8 OBLAST

Title:

Osteoblastic cells in weightlessness: morphology and biochemical response **Experiment identifier:** BIOK-88-12-F **Study object:** ROS 17/2.8 (osteoblastic cell line) **Investigators:** C. Alexandre, LBTO, Faculté de Médécine, Saint-Etienne (F) C. Genty, LBTO, Faculté de Médécine, Saint-Etienne (F) S. Palle, LBTO, Faculté de Médécine, Saint-Etienne (F) **Funding:** ESA and French national funding (CNES)

9.8.1 Summary

Monolayers of osteosarcoma cells were grown in space from pre-confluence to confluence. After four days, the shape of the cells began to change, culminating after six days in a mixture of morphologies that included contracted cells with cytoplasmatic extensions and rotund cells layered on top of each other. A normal cell morphology was displayed in the 1*g* controls, both on ground and in flight. From the six-day cultures, double the alkaline phosphatase activity was extracted than from the corresponding ground controls. The rate of cell proliferation was not affected.

9.8.2 Scientific objectives

Bone mass loss in space can either be explained by an enhanced activity of bonedegrading cells (osteoclasts) or by a reduced activity of bone forming cells (osteoblasts). A combination of both is also conceivable. In the O(STEO)BLAST experiment a closer look was taken at the behaviour of osteoblasts in weightlessness. Osteoblastic cells were cultured *in vitro* to eliminate the many external stimuli that are thought to influence the cells *in situ* under weightlessness: unloading of the skeleton, redistribution of body fluids, altered hormone levels and so on.

9.8.3 Biological materials

Freshly-prepared osteoblasts were initially considered for the OBLAST experiment. When pre-flight tests indicated that the quality of such primary cultures was too variable, clonal cells (ROS 17/2.8, an osteogenic cell line derived from rat bone carcinoma) were chosen as a more dependable alternative. The cells were grown as monolayers on plastic cover slips (size: $22 \times 9.5 \times 0.2$ mm) at an initial density of 5×10^3 cells/cm² (pre-confluent).

9.8.4 Experiment hardware

OBLAST was one of the four experiments in Biobox. As was the case for the other Biobox experiments (BONES, MARROW and FIBRO) the hardware consisted of the experiment units described in Section 6.3.1. The main material used to construct the units of the OBLAST experiment was PSU (polysulphone), a semi-transparant, yellowbrown plastic. Most OBLAST plunger units carried one cell culture although some carried two (Table 9-10). The OBLAST plunger units were hermetically closed with no provision for gas exchange.

9.8.5 Chemicals and materials

Culture medium

The cells were cultured as a closed system, without CO_2 supply. The medium consisted of:

- DMEM (Dubelco's Modified Eagle's Medium);
- 20mM Hepes buffer with 1% NaHCO₃ (a dual buffer);
- 10% bovine fetal serum;
- L-glutamine (292 mg/l);
- streptomycin (100 mg/l);
- penicillin (100,000 U);
- kanamycin sulphate (100 mg/l).

The pH value of the medium was between 7.2 and 7.4

Cell supports

The cells were grown on cover slips made of Thermanox plastic.

Buffer

The cultures were rinsed in PBS at 1N before being fixed or solved.

Figure 9-13. Microgravityinduced changes in the shape of osteogenic cells. Left: normal cell morphology (flat polygons) as observed at 1g. Right: contracted, bulbous cell with cytoplasmatic extensions as found in microgravity. The OBLAST and FIBRO experiments on Bion-10 were among the first to report microgravity-induced morphological changes in mammalian cell cultures.





Fixative

The fixing solution was 2% glutaraldehyde in sodium cacodylate buffer at 0.175 Mol/l, pH 7.1.

Lysing solution

The lysis solution consisted of 0.1% Triton X-100 in Tris (1.21 g/l).

9.8.6 Mission timeline events

The cell cultures were loaded into the experiment units at LBTO in St-Etienne (F), five days before launch. The units arrived in Moscow four days before launch. The active phase of the OBLAST experiment spanned the first six days of the flight. The cell cultures were sequentially fixed in four groups, after

Table 9-10.	Experiment units	Cell cultures
Flight microgravity	n= 7	n= 28
Flight 1g (centrifuge)	n= 2	n= 2
Ground 1g	n= 7	n= 9
Total	n= 16	n= 20

being exposed to microgravity for 0, 2, 4 or 6 days. Lysis was performed solely at day 6.

9.8.7 Results

Technical performance

All 16 OBLAST plunger units (Table 9-10) worked well. All 84 fluid transfers were executed as planned. One culture out of 20 was lost due to culture medium leaking from the plunger unit.

Impact of the thermal anomaly and early return of the spacecraft

The thermal anomaly in Biobox (described in Section 6.6) occurred when the active phase of the OBLAST experiment was over, with all cell cultures chemically fixed or dissolved. The uncontrolled storage temperature in Biobox by the end of the flight did not significantly reduce the scientific return of OBLAST, as the experiment was designed anyway to see high temperatures after sample fixation. The early return of the capsule as such had no impact on OBLAST.

Table 9-10. Sample configuration of OBLAST. Most plunger unit carried one, some carried two cultures.

Scientific results

The rate of proliferation was measured by determining the cell numbers after 0, 2, 4 and 6 days. The cell density almost doubled during this period, both in microgravity and at 1g, with the cultures reaching confluence.

In the cultures exposed to microgravity, a change in cell shape was observed from day 4 onwards, being overtly displayed on day 6. While at 1g the cells maintained their angular, stretched-out morphology throughout the experiment, the cells in microgravity developed a mixture of appearances, ranging from the normal flat shape to strongly contracted cells with cytoplasm extensions running along the Thermanox surface (Figure 9-13), and bulbous cells defying the monolayer arrangement by piling on top of each other. The changes in cell shape indicated that the cytoskeleton was somehow affected. An analysis of the cells with a transmission electron microscope did not reveal any other changes at the ultrastructural level, however. While the total protein content of the microgravity and 1g cultures was the same at day 6, the microgravity cultures contained about double the alkaline phosphatase activity, suggesting that the cell activity changed along with the changes in morphology. In addition, indications were found that less osteocalcin had been produced by the microgravity cultures.

Using the same type of cell cultures and experiment units as premièred on Bion-10, the OBLAST investigations were continued on the Zero-g Caravelle in ESA's 18th and 21th Parabolic Flight campaigns (1994, 1995) and in Biobox flown on Foton-10 (1995) and Foton-11 (1997).

Since the flight of the OBLAST experiment on Bion-10, microgravity-induced cell shape changes in *in vitro* cultures of osteoblasts have been observed by other investigators as well. This phenomenon is nowadays considered an accepted fact [11]. In contrast, the reported increase of alkaline phosphatase could not be reproduced, neither by the OBLAST team, nor by other investigators, and remains therefore debatable.

9.8.8 Publications

Genty C., Palle S., Alexandre C., 1993. First osteoblast culture in space: cellular response to unloading. *Journal of Bone and Mineral Research*, **8**: Suppl.1, p S367.

Genty C., 1993. Réponse *in vitro* des cellules ostéoformatrices aux stimuli mécaniques. Etude en microgravité réelle et sur des modèles animaux après surcharge et décharge mécaniques. PhD Thesis, Université Jean Monnet, Saint-Etienne, France, pp 130-148.

Guignandon A., Genty G., Vico L., Lafage-Proust M.-H., Palle S., Alexandre C., 1997, Demonstration of feasibility of automated osteoblastic line culture in spaceflight. *Bone* **20**:2, pp 109-116.

Vico L., Lafage-Proust M.-H., Alexandre C., 1998. Effects of gravitational changes on the bone system *in vitro* and *in vivo*. Bone **22**:5, pp 95S-100S.

9.9 SEEDS

Title:

Biological effects by cosmic heavy particles including protons in Arabidopsis seeds **ESA experiment identifier:** BIOK-88-11-D (second reflight) Study object: Plant seeds (species: Arabidopsis thaliana) **Investigators:** A.R. Kranz, J.W. Goethe-Universität, Frankfurt-am-Main (D) B. Baican, J.W. Goethe-Universität, Frankfurt-am-Main (D) K.E. Gartenbach, J.W. Goethe-Universität, Frankfurt-am-Main (D) E. Schopper, J.W. Goethe-Universität, Frankfurt-am-Main (D) M.W. Zimmermann, J.W. Goethe-Universität, Frankfurt-am-Main (D) J.-U. Schott, DLR, Cologne (D) C. Heilman, CRN, Strasbourg (F) R. Selz, CRN, Strasbourg (F) V.V. Shevchenko, Koltzov Institute of Developmental Biology, Moscow (RUS) A.M. Marenny, RCSRS, Moscow (RUS) V.E. Dudkin, RCSRS, Moscow (RUS) Yu.V. Potapov, RCSRS, Moscow (RUS) N.A. Nefedov, RCSRS, Moscow (RUS) **Funding:**

ESA and German national funding (DARA)

9.9.1 Summary

Seeds of Arabidopsis thaliana, accompanied by track detectors, were exposed to space radiation to determine to what extent the proton segment of the radiation spectrum contributes to the biological damage observed in plant seeds after spaceflight. This question remained unresolved because the SEEDS team broke up before the postflight analysis was completed.

9.9.2 Scientific objectives

The objective of SEEDS was to investigate how, and to what extent, plant seeds are damaged when hit by cosmic particles. The experiment flew consecutively on Bion-8, -9 and -10. On Bion-8, a considerable amount of biological damage was found that could not be correlated with the impact of cosmic heavy

ions as recorded by the plastic detectors. (A similar problem was encountered in the WOLFFIA experiment described in Section 9-10.) Therefore, on Bion-9 the experiment was repeated with different detectors of higher sensitivity. Even then, it seemed that undetected, low-LET particles had significantly contributed to the biological damage. The objective of flying SEEDS on Bion-10 was to pay special attention to these low-LET particles, i.e. the proton segment of the radiation spectrum (LET values down to 100 MeV per cm H_2O (= 10 keV per μ m H_2O)).

The Arabidopsis seeds were stacked between the radiation detectors, allowing hit and nonhit individuals to be discriminated after flight by correlating their spatial location with the tracks left by the particles. After flight, the radiation damage was quantified by measuring the percentage of non-germinating seeds and, in those seeds capable of germinating, by measuring subsequent developmental aberrations. By using well-defined genetic lines of Arabidopsis, it was envisaged to track down the aberrations to specific gen deletions in the seeds chromosomes.

9.9.3 Biological materials

The flight set of SEEDS contained 22200 dry seeds of the crucifer plant Arabidopsis thaliana Heynh (L.), diploid wild type En-2 including one-gene marker lines of the five chromosomes, each designed for localising induced gene mutations in subsequent studies. The oval seeds, about 0.5 mm in length, were glued on glass plates at an approximate density of one seed per mm². The plates were interleaved with track detectors following the Biostack concept [12]. Enclosed inside the stack, the seeds were maintained in dry air.

9.9.4 Experiment hardware and materials Apart from minor modifications, the hardware used on Bion-10 was identical to the SEEDS hardware flown on Bion-9 [8]. The detectors came in two kinds: nuclear photo emulsions and silver halide (AgCl) detectors.

Photo emulsions: SEEDS 1a and 1b The photo emulsions were installed in two Biorack Type I containers, labelled SEEDS 1a and SEEDS 1b (Figure 9-14). The detectors consisted of a 0.5 mm-thick glass plate, coated on both sides with a 0.2 mm-thick Ilford K2 nuclear photo-emulsion. On either side of the detector, a 0.17 mm-thick glass plate with 700 glued-on seeds was mounted. Each container carried six of these detector and seed sandwiches.

The two containers were placed in two KNA facilities mounted on the external surface of the spacecraft where they would receive a maximum dosage of space radiation (see Chapter 7). To dampen the temperature oscillations caused by the solar illumination and shadowing cycles in orbit, SEEDS 1a and 1b were covered by a multi-layer insulating (MLI) hood, manufactured by ESA. Celsistrips were included to record the maximum temperature experienced during flight. SEEDS 1a and SEEDS 1b were identical, apart from the 1 mm-thick steel plate that was mounted in front of the latter to form a well-defined radiation shield.

Silver halide: SEEDS 2

The silver chloride detectors, with ancillary equipment, were accommodated in a more roomy Type II container, labelled SEEDS 2 (Figure 9-4). Each detector consisted of a 1 mm-thick glass plate, coated by a 140 µm monochrystalline layer of cadmium-doped silver chloride. On one side of the detector a 500 µm macrolon plate was mounted with 1800 gluedon seeds. The container carried three such sandwiches. The three-storey detector and seeds stack was illuminated by a 49-piece array of yellow light-emitting diodes to stabilise the tracks in the silver chloride detectors. The lightemitting diodes and their power supply (four lithium batteries) were encased in the same container. The total number of seeds in SEEDS 2 was 5400 (three plates of 1800 seeds each).

SEEDS 2 was placed in a Russian BB box, shared by WOLFFIA, DOSICOS 5 and other experiment packages. This BB box was mounted on the floor panel of the Bion capsule (Figure 2-7). In this location, they must have been unintentionally exposed to the rays of the onboard gamma source, described in Chapter 5.



9.9.5 Mission timeline events

8400 seeds

The SEEDS containers were integrated on 4 December 1992, at the University of Frankfurt. After hand-over to ESA, the packages were flown to Moscow on 15 December. Transport of the SEEDS flight set from Moslab to Plesétsk took place on 22 December (SEEDS 1a and 1b) and 26 December (SEEDS 2). The electrical power of SEEDS 2 was switched on in Moscow just before departure to Plesétsk. SEEDS was integrated with the spacecraft on 27 December at L-2 days.

8400 seeds

During flight, ground controls of SEEDS 1a and 1b were stored in Moslab at ambient temperature, while the control of SEEDS 2 underwent a 24-h delayed simulation of the spacecraft temperature in a thermal chamber in IBMP's Building 4.

SEEDS 1b, one of the two external packages, was planned to be analysed by IBMP. After a preliminary post-flight inspection in Frankfurt, it was handed over to IBMP on 31 August 1993 during the data exchange meeting in Kiev. Figure 9-14. Configuration of the SEEDS experiment on Bion-10.

LEDs + batteries 5400 seeds
9.9.6 Results

Impact of the thermal anomaly and early return of the spacecraft

The temperature profile recorded by NASA in KNA-1 showed spikes up to 60°C during the second half of the flight, when the spacecraft flew out of eclipse (Figure 7-12). Nevertheless, the maximum temperature inside SEEDS 1b as indicated by the Celsistrips never exceeded 40°C, which suggests that the MLI hood effectively protected the experiment against severe thermal transients. SEEDS 2, located inside the capsule, was subjected to temperatures up to 33°C during the second half of the flight. These figures would suggest that in SEEDS 1b and SEEDS 2 no temperatures were experienced that could have harmed the seeds or the detectors.

The early return of the spacecraft caused SEEDS to be exposed to space radiation for 11.6 days instead of 13.6 days, but this reduction did not essentially affect the experiment.

Thermal damage during re-entry

After flight, a dark streak was found on the MLI hood of SEEDS 1a, indicative of exposure to high temperatures. The change of colour was confined to the outmost layer of the hood. The highest temperature (estimated) at the outer surface must have been around 100°C. As SEEDS 1a was located in KNA-2, whose lid was not properly closed at the end of the flight, the streak was apparently caused by hot air streaming across the hood of SEEDS 1a during re-entry. The maximum temperature recorded by the Celsistrips inside SEEDS 1a was in the range 77-81°C. Many seeds in SEEDS 1a appear to have been killed by the high temperature (see below).

Dosimetric results

Outside the capsule (SEEDS 1b, shielding ~1 g/cm²) the recorded dose rate was 870 ± 44 µGy per day, of which 7 µGy per day (less than 1%) was attributable to heavy ions (LET > 10 keV/µm). As expected, inside the capsule (SEEDS 2, shielding ~15 g/cm²) the dose rate was much lower. A value of 174 ± 4 µGy per

day was measured, with 2.6 μ Gy per day attributable to heavy ions (LET > 10 keV/ μ m), the flux of which was 168.5 particles per cm² per day.

Biological results

In SEEDS 1a (the overheated package) most seeds had died; the germination rate in comparison to the ground control was only 14.3%. The germination rate of SEEDS 1b (the non-overheated package) was 50.3% of the rate in the ground control. From SEEDS 2, 76.8% of the seeds was able to germinate after flight.

Unfortunately, the post-flight fate of the individual seeds was never correlated with the recorded particle tracks. The post-flight analysis came to a premature stop for a variety of reasons, as summarised below.

Financial support for the experiment was not continued during 1993 after the Principal Investigator, Prof. A.R. Kranz, retired in April 1993 and left his laboratory at Frankfurt-am-Main, three months after the (delayed) space flight of Bion-10. Somewhat later, K.E. Gartenbach and M.W. Zimmermann changed their jobs. B. Baican, who was engaged with the dosimetry and responsible for track measurements, fell seriously ill. V.E. Shevchenko and V.V. Dudkin, whose participation in SEEDS on Bion-8 and -9 was partly supported by western sources, were unable to continue when funding could not be extended. Meanwhile, the Russian financing of space experiments by RCSRS had come to an end, forcing A.M. Marenny, Yu.V. Potapov and N.A. Nefedov to stop work on SEEDS.

9.9.7 Scientific cooperation

The experiment packages were prepared by the ESA-affiliated investigators. After flight, one of the two outside-exposed packages was planned to be analysed by the IBMP-affiliated investigators. Tasks were distributed as follows:

Biology:

A.R. Kranz, K.E. Gartenbach, M.W. Zimmermann, V.V. Shevchenko *Plant seed preparation and detector hardware construction:* J.-U. Schott

AgCl detectors, emulsions, microdosimetry: E. Schopper, B. Baican, J.-U. Schott

Particle track analysis of the emulsions: C. Heilman, A.M. Marenny, V.E. Dudkin, N.E. Nefedov, Yu.V. Potapov

9.9.8 Publications

Despite the unfortunate developments in the latter stage of the SEEDS experiment, several publications were produced.

The dosimetric data and the seeds' survival rates have been published in combination with earlier findings from Cosmos 2044 (= Bion-9) by the SEEDS team.

Gartenbach K.E., Zimmermann M.W., Kranz A.R., Baican B., Schopper E., Schott J.-U., Shevchenko V.V., 1994. *Present results of the joint radiobiological ESA - IBMP experiments "SEEDS" aboard Cosmos 2044 and 2229 – correlation of microdosimetric data and different biological endpoints in* Arabidopsis thaliana. Book of Abstracts, 30th Cospar Scientific Assembly, Hamburg, Germany, 1994, p. 300.

Gartenbach K.E., Kranz A.R., Zimmermann M.W., Schopper E., Baican B., Schott J.-U., Heilmann C., Schevchenko V.V., 1996. Present results of the joint radiobiological ESA – IBMP experiments "SEEDS" aboard Cosmos 2044 and 2229 – correlation of micro-dosimetric data and damage endpoints in *Arabidopsis thaliana* plants. *Adv. Space Res.* **18**:12, pp 215-220.

In addition, a comparison of the results obtained with experiments flown on the SL-1, LDEF, IML-1, D-2, ERA-1, Bion-8, -9, -10 missions has been published:

M.W. Zimmermann M.W., Gartenbach K.E., Kranz A.R., Baican B., Schopper E., Heilmann C., Reitz G., 1996. Recent results of comparative radiobiological experiments with short and long term expositions of *Arabidopsis* seed embryos. *Adv. Space Res.* **18**:12, pp 205-213.

9.10 WOLFFIA

Title:

Radiation damage in *Wolffia arrhiza* caused by heavy ions of cosmic rays **Experiment identifier:** (spin-off from) BIOK-88-9D **Study object:** Duckweed (species: *Wolffia arrhiza*) **Investigators:** G. Reitz, DLR, Cologne (D) R. Facius, DLR, Cologne (D) L.V. Nevzgodina, RSSS, Moscow (RUS) Ye.N. Maksimova, RSSS, Moscow (RUS) Yu.A. Akatov, RSSS, Moscow (RUS) **Funding:** Russian (IBMP) and German national funding (DLR)

9.10.1 Summary

WOLFFIA was an investigation into the longterm post-flight effects of space radiation on plants. Following the Biostack concept, duckweed (species: *Wolffia arrhiza*) was sandwiched between plastic radiation detectors. After flight, the plants were subcultured and monitored for three months. An enhanced rate of mortality was observed in the flight series, accompanied by an increased percentage of morphological anomalies. The damage could partially be linked to the passage of heavy ions through the plants.

9.10.2 Scientific Objectives

The duckweed *W. arrhiza* was a new species to the vast Biostack programme, whose radiobiological experiments had been conducted under the aegis of the German Aerospace Centre, DLR, and its predecessor DFVLR, since 1972 [12]. Whereas investigations of desiccated, dormant life-forms like spores, cysts and seeds have dominated Biostack, "wet" specimens were occasionally flown as well, such as insect eggs in the CARAUCOS experiments flown on Bion-8 and -9 and duckweed in the WOLFFIA experiment on Bion-10.

HZE radiation acts differently on dry and wet organisms. In a wet system, the cosmic rays act on the biomolecules not only directly, but also indirectly through water radiolysis.



Figure 9-15. The WOLFFIA experiment package was configured as a traditional Biostack, with five biolayers carrying Wolffia plants interleaved between 6 × 3 detector foils of cellulose nitrate (CNk). The biolayers came in two varieties, referred to as type 1 and type 2 (see text). Each green dot represents an individual Wolffia plant. Another difference is that the cells in a wet, metabolising, organism are capable of repairing radiation damage, a feature that is not found in dehydrated, dormant forms of life such as seeds, spores or cysts. Wet test systems are particularly relevant to radiation hazard studies in support of manned spaceflight because of the large amount of water present in the human body.

9.10.3 Biological materials

The subject of the study was the duckweed *Wolffia arrhiza*, the world's smallest flowering plant. The size of an adult *Wolffia* measures just 1.5 mm. This 'mini-plant', which is found floating on the surface of fresh water ponds, consists of a single bead-like leaf without stem or root (*arrhiza* is Greek for root-less). Alternatively, some textbooks describe *Wolffia* as a stem without leaves or roots.

W. arrhiza reproduces predominantly vegetatively by budding. The dorsal side of the leaf bears a basal pocket with meristematic cells in which the bud is formed and out of which a new leaf develops, ultimately splitting off after some time. As the new leaf is genetically identical to its parent, W. arrhiza can be said to produce clones of itself. Having delivered up to seven such clones, the parent plant dies. Wolffia can stay alive for at least one month in darkness if maintained under humid conditions. This capability was exploited in the WOLFFIA experiment by storing the plants on moistened filter paper inside the detector package. With their metabolism suppressed - no growth, no development, no reproduction - the plants

survived the confinement by living off their reserves.

9.10.4 Experiment Hardware

The WOLFFIA experiment package was configured as a traditional Biostack, with five biolayers carrying *Wolffia* plants, interleaved between 18 detector foils of cellulose nitrate. (Figure 9-15). The biolayers came in two varieties, referred to as type 1 and type 2:

Biolayer type 1

This biolayer was designed for hit/non-hit assessments in individual plants. It comprised a Makrolon plate measuring $80 \times 40 \times 3$ mm, in which 20 furrows had been cut. Each furrow was lined with moistened synthetic sponge material, carrying up to 15 *Wolffia* plants on top. Each type 1 layer carried between 240 and 270 plants.

Biolayer type 2

Densely-packed, multilayered *Wolffia* plants were subjected to gross radiation assessments at the population level. Makrolon plates measuring $80 \times 40 \times 3$ mm were covered with moistened filter paper and populated with 70 individuals per cm², approximately 1000 plants per layer.

The *WOLFFIA* stack was encased in an ESA Biorack Type I container which was placed in an upright position inside a Russian BB Box, together with SEEDS 2, DOSICOS 5 and other experiment packages. The BB Box was mounted on the floor panel of the Bion capsule (Figure 2-7).

9.10.5 Materials

The track detectors foils were made of cellulose nitrate produced by Kodak (CNK). The biolayers were made of polycarbonate (trade name: Makrolon).

The type 1 biolayers were lined with moistened plastic sponge to prevent the plants from drying. Similarly, the type 2 biolayers were lined with moistened filter paper to prevent the plants from drying. The 23-layer stack was held together by an aluminium frame.

9.10.6 Mission timeline events

The duckweeds were integrated with the Biostack one week before launch and removed from the Biostack one week after landing, bringing the total stay inside the stack to about 3.5 weeks. The flight experiment was accompanied by a synchronous control experiment on ground in a duplicate Biostack, likewise encased in a Type I container.

9.10.7 Results

Effect of the thermal anomaly and early return of the spacecraft

The high temperatures may have promoted the development of fungal infections inside the stack (see below). It may also have aggravated the stress experienced by the plants inside the Biostack, exemplified by the high background rate of anomalies observed in the ground controls. The experiment was not essentially affected by its premature return to Earth.

Hit versus non-hit

After flight, fungal infections were found in the Biostack. The presence of fungi considerably reduced the number of plants available for analysis. Eventually, only one of the three type 1 biolayers was investigated in full detail. This biolayer had been traversed by 381 heavy ions (detection threshold: Z>8). From the 242 *Wolffias* in this layer, 41 plants (17%) were positioned in the path of such a particle, of which six (2.5%) had been hit in the bud.

Mortality

The 47% death rate in flight was higher than that observed on ground, which was 27%. Still, the mortality of the plants that had been hit was not specifically enhanced (no positive correlation could be found). The low rate of

survival may have been due to a variety of flight factors, including low-LET HZE radiation not registered by the track detectors and gamma radiation from the on-board source (see Section 5.1).

Out of the six plants that had been hit in the bud, five were found to be dead. Although the sample size was small, this suggested that these five plants were actually killed by the collision with a heavy particle, whose damaging effect seemed to be particularly lethal when localised in the plant's reproductive organ, i.e. in the budding zone.

Morphological anomalies

Morphological anomalies of many kinds have been described for *W. arrhiza*. Examples are twin buds sprouting from a single parent, offspring that remain attached to the parent plant, out-of-size plants (dwarfs and giants) and so on. Usually, the fraction of anomalous individuals in a given population is less than 1%.

After flight, anomalies were observed in about 3% of the plants, in the flight and ground series alike. After 1-2 weeks of post-flight culturing this fraction had grown to 13% for the flight samples and 8% for the ground controls. The steep rise may be ascribed to the emergence of a new generation, which had undergone the stress in the Biostack while still in the bud stage of their development. The higher percentage observed in the flight series would illustrate that the conditions during spaceflight were more stressful than on ground (cf. the enhanced rate of mortality).

In the period that followed, the difference between the flight and ground series gradually disappeared. With the cultures proliferating exponentially, the percentage of anomalies gradually fell off in both groups, returning to its original value below 1% after three months of post-flight culturing.

Since the lifetime of a *Wolffia* individual is of the order of one-and-a-half months, this suggests that in the latter half of the threemonth period excess anomalies were displayed by a generation that was never stressed by the Biostack conditions. If so, the anomalies must have been passed on by parents whose bud region was irreversibly damaged. To investigate whether the anomalies were due to the passage of heavy ions through the plants, hit and non-hit *Wolffias* taken from the flight series were separately subcultured. More anomalies were indeed found in progeny produced by parents that had been hit, but the small sample size precluded ultimate proof of this point. It is worth noting that the cultures were back to normal after three months. Apparently, the anomalous individuals ultimately lost the competition for survival, which was won by the normal individuals.

9.10.8 Scientific cooperation

The ESA investigators were responsible for the Biostack hardware, the dosimetry and the hit/non-hit analysis. The IBMP investigators took care of the biological part of the experiment (in practice, everything related to the *Wolffia* plants).

9.10.9 Publications

Nevzgodina L.V., Maximova E.N., Kaminskaya E.V., 1994. *Biological effects induced by heavy ions in Wolffia arrhiza higher plants exposed onboard Biosatellite-10*. Book of Abstracts, 30th Cospar '94 Scientific Assembly, Hamburg, Germany, 1994, p 300.

Maksimova E.N., Nevzgodina L.V., Kaminskaya E.V., Akatov Yu.A., Arkhangelskij V.V., 1994. *Radiobiologícheskie effékty na populyátsii Voľ fii v kosmícheskikh i nazémnykh eksperiméntakh* (*Radiobiological effects on populations of Wolffia in space and ground-based experiments* [in Russian]). Abstracts of the Xth Conference Space Biology and Aerospace Medicine, p 282, Moscow.

Facius R., Scherer F.K., Strauch W., Nevzgodina L.V., Maximova E.N., Akatov Yu. A., 1996. Mortality and morphological anomalies related to the passage of cosmic heavy ions through the smallest flowering aquatic plant *Wolffia arrhiza*. Advances in Space Research, **18**:12, pp 195-204.

9.11 Scientific Return

9.11.1 Summary of the output of the ten experiments

Of the ten experiments jointly flown by ESA and IBMP on Bion-10, ALGAE (Section 9.1) was a direct victim of the early return of the spacecraft. Another experiment, CLOUD (Section 9.3), failed because of a technical mishap. For a third experiment, SEEDS (Section 9.9), the biological analysis was never completed. The remaining seven experiments yielded significant results.

The dosimetrical study DOSICOS (Section 9.4) delivered some good results despite the damage suffered by some of the detectors through overheating. The ethological insect study FLIES (Section 9.6) produced intriguing new information and enough raw material to generate a string of follow-on experiments in space. The promising radiobiological experiment WOLFFIA (Section 9.10) would have deserved a similar success but this experiment was unfortunately never reflown.

The most interesting results were probably obtained from the four Biobox experiments, BONES, FIBRO, MARROW and OBLAST, (Sections 9.2, 9.5, 9.7 and 9.8 respectively), each aimed at investigating the effects of microgravity on mammalian cells and tissue cultures in vitro. These four studies probed into scientific regions that were still uncharted in 1992. The added value of the Biobox output was that the results of the four investigations complemented each other very well [13]. By comparing the four sets of data (each of them already worthwhile in their own right), a set of patterns and trends became visible that has maintained its validity up to the present day. The commonality and consistency in the results of BONES, FIBRO, MARROW and OBLAST can be summarised as follows:

- (i) All culture types reacted some way to the absence of gravity (BONES, MARROW, OBLAST and FIBRO).
- (ii) In none of the cultures was a difference in growth rate detected:

- the incorporation of thymidine was unchanged (FIBRO);
- the protein content was unchanged (MARROW, OBLAST);
- the DNA content was unchanged (MARROW);
- the overall growth was unchanged (BONES);
- the cell number was unchanged (OBLAST).
- (iii) All reported effects were somehow associated with changes in cell activity:
- reduced production of collagen type 1 (MARROW);
- reduced production of osteocalcin (OBLAST);
- reduced production of alkaline phosphatase (MARROW);
- reduced rate of mineralisation (BONES);
- enhanced production of collagen type 1 upon stimulation by IL-1 (MARROW);
- enhanced production of alkaline phosphatase upon stimulation by PTH (MARROW).
- (iv) In all (non-stimulated) osteogenic cultures, the bone-forming activity was suppressed (BONES, MARROW, OBLAST).
- (v) The four observed effects (i, ii, iii, iv) seemed to be independent of the level of biological organisation, as they were found in isolated organs (BONES), threedimensional cultures (FIBRO, MARROW) and monolayer cultures (FIBRO, OBLAST).
- (vi) In those cultures that were configured as monolayers (and not in any other), changes in cell morphology were observed:
- mixed morphologies, including more contracted, rotund cell shapes (OBLAST);
- more contracted, rotund cell shapes (FIBRO).

9.11.2 Biobox results under discussion

The phenomenon of bone mass reduction in space is thought to originate at the cellular

level, where the bone mass budget is controlled by the antagonistic efforts of osteoblasts (bone-forming cells) and osteoclasts (bone-degrading cells). However, it is not known how these cells are triggered to change their activity in space: the osteoclasts may feel the loss of tension in the weightcarrying bones in space ('unloading of the skeleton'), stress hormones released in the body could play a role, the re-distribution of body fluids in weightlessness could have an impact and, finally, it could be that osteoblasts and osteoclasts can detect the absence of gravity directly. To test the last hypothesis, in vitro cultures were used in Biobox, consisting of cells that were literally cut off from any signals generated by the body [13].

Prior to the flight of Biobox on Bion-10, very little had been accomplished in understanding the behaviour of bone-forming cells *in vitro* under microgravity conditions [14]. In retrospect, the Biobox experiments on Bion-10 may be considered as the first systematic set of investigations in this field. Since then, similar and complementary investigations have been carried out on Foton capsules (facilities: Biobox, Ibis) and on the US Space Shuttle (facilities: Biorack, Biobox, Osteo and others) with results often in agreement with the trends discovered in 1993 and listed in the previous paragraph.

The collective results from these experiments seem to support the idea that bone-forming cells may react autonomously to the absence of gravity, without necessarily being signalled by the body. For many years, however, arguments have been adduced by biophysicists against this concept [15-20]. Their arguments may be summarised as follows:

(a) From a biophysical point of view, gravisensing by a single cell is impossible owing to its small size. Within a cell, the forces generated by weight are considered too minute to produce any effect, unless supported by some intracellular amplifier or gravisensor. However, no amplifier or gravisensor has so far been found (see b);

- (b) Neither specialised gravisensing organelles, nor any alternative gravisensing mechanism has ever been identified in a mammalian cell (in contrast, such sensors *have* been found in plant root cells and in some protists);
- (c) The reported effects could well be *indirect*: the lack of convection in the culturing fluids under microgravity may have an impact on the activity of the cells (Note: proof of this alternative explanation is still missing).

At the time of writing, the question of whether, and if so, how, bone cells can detect the presence of microgravity directly is still hotly debated [21-25]. Therefore, it becomes increasingly urgent to design and perform new experiments that can bring clarity to this controversy.

9.11.3 The relevance of the Biobox experiments to osteoporosis

The rapid loss of bone mass during spaceflight may be used as an appropriate test model for the loss of bone in elderly people on ground (osteoporosis), which is nowadays considered to be a major medical – as well as financial and economic – problem. Future investigations in space into the behaviour of bone-forming cells in microgravity will partly be funded by the pharmaceutical industry under ESA's Microgravity Applications Programme (MAP). As such, Biobox has helped to lay the basis for an important branch of medical research in space with a terrestrial application.

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10. The Post-Bion-10 Era

On Bion-10, as previously on Bion-8 and Bion-9 [1], IBMP acted as the interface between ESA and Russian organisations. As a research institute, IBMP was not ideally equipped with the managerial tools needed to operate as a payload integration organisation even at the scale of Bion-10. The lack of experience in this field was aggravated by the effects of the crisis the country went through during the years described in this report. In particular the social instability, hyper-inflation [2] and concerns about job security exerted a strong influence. Some of the unfortunate events that occurred in the interaction between ESA and IBMP can be attributed to these adverse conditions, but others would not have run to the same extent in organisations more oriented towards engineering and operations. This comment particularly refers to issues of transport, logistics and communications during all phases of the campaign.

IBMP also turned out to be less involved in the details of the spacecraft design and its operation than was anticipated in ESA. Hence, we were insufficiently informed about features like the spacecraft system's heat rejection capabilities, its sensitivity to out-of-eclipse conditions, the onboard gamma-ray source and the human and data interface with the Flight Control Centre (TsUP). As a result of all of this, there was a real risk that IBMP could end up in an unfavourable position for a candidate future mission manager for complex ESA life science payloads.

The post-Bion-10 era began long before Bion-10 itself was launched. As early as January 1992, IBMP had invited ESA and other international partners to a meeting in Moscow to discuss the future of the Bion programme. The huge economic problems caused by the

ongoing disintegration of the Soviet Union were a direct threat to the financing of the country's space programme. In this context, the traditional cooperation with IBMP, based on a policy of no-exchange-of-funds could not survive. From now on, international participation in the Bion programme could be realised only on a cost-reimbursement basis. During the meeting, which took place in February 1992, an association for the promotion of life science research in space was proposed by IBMP, whose membership included the Institute itself and two of the key companies in Russia involved in the Bion programme. This association was to become the main shareholder of a commercial superstructure, which would include some recently created Moscow-based joint ventures to deal with the financial and legal aspects of



ESA payload mass on Bion/Foton (kg)

Figure 10-1. Payload mass flown by ESA on Bion-Foton spacecraft since 1987. Foton-7 is not included because ESA's contribution to this mission consisted only of protein samples (Russian-provided hardware was used). ESA's payload mass figure for Foton-M1 is an estimate, as the mission was not fully defined at the time of writine. foreign contracts. At this time, IBMP intended to develop a Bion-*B* spacecraft for general biology research, to be launched in 1993, a Bion-*R* for rodent investigations to be launched in 1994, and a Bion-*P* for primates, to be launched in 1995. ESA's involvement would, almost exclusively, be in Bion-*B*, with the Biobox and Biopan facilities.

Considering the disarray in which the country found itself during the whole of 1992, this programme was far too ambitious. Within a month, IBMP had changed its commercial partners, and throughout the year uncertainties prevailed as to the solidity of the business setup. In addition, at the end of June 1992 the newly established Russian Space Agency (RKA) informed ESA that public funding of Bion-B was not guaranteed. The gap to be bridged by money gained from contracts with the West now became too wide for IBMP's *kommersanty* (business representatives), who soon thereafter disappeared from the scene.

During the latter part of 1992, IBMP tried a different approach. This time a stock company called BION was put forward, with IBMP itself, TsSKB from Samara and one local commercial company as its shareholders. At this time the forecast launch date of Bion-B had already slipped to August 1994, and Bion-R and Bion-P had vanished altogether. Instead, a Bion-11 mission carrying two monkeys in orbit was now contemplated for December 1994.

By the middle of 1992 there were also intensified efforts by industry in western Europe to obtain direct contracts from the Foton-Bion spacecraft manufacturer in Samara for marketing flight opportunities in ESA member states. It was during this period that the exclusive attention to *Bion* changed in favour of a more general focus on *Foton-Biontype* spacecraft. In fact, both spacecraft were produced by TsSKB in Samara and they were largely similar.

Various European companies were involved in marketing the Foton-Bion activities, and they combined their resources in October 1992 while ESA and other organisations were still negotiating with the BION company. At one point, there were three different factions within one and the same Russian company negotiating the same subject separately and against each other with different bodies in ESA member-states. In February 1993 ESA definitively rejected any brokerage by industry for its flight opportunities on Foton-Bion and continued its discussions only with RKA and IBMP.

In the meantime, ESA had gained its first experience with the Cosmos Group of Samara, comprising TsSKB and partners, which had integrated the Biopan instrument on Foton-8 for its maiden flight, although it was initially designed as an element of cooperation between ESA and IBMP for accommodation on Bion missions. Despite some serious misgivings about the flight operations, considerable advantages had been noted in the areas of ground operations and payload integration. It was becoming clear that, unless IBMP's BION company came forward with a convincing proposal, an engineering organisation like the Cosmos Group would win the competition. This is exactly what happened in the second half of 1993 (see below).

Shortly after the completion of the Bion-10 mission, engineering and operations debriefings took place. In the area of *flight operations* several conclusions were drawn. First of all, because of the demonstrated sensitivity of the spacecraft to out-of-eclipse conditions, detailed advance information about the planned orbital parameters would be required by ESA in the future. Both Bion-9 and Bion-10 suffered from heat rejection problems during the last days of their missions.

Secondly, ESA would not only continue to insist on near-real time telemetry data from the TsUP, but worked out an overall set of rules to determine the interaction between the TsUP and the payload community.

Also, a very preliminary scientific balance of the mission was drawn up. One of the more *science*-oriented lessons learned led to the

Table 10-1

Mission	Year	ESA multi-user facilities		acilities	Flight opportunity offered by
		Biobox	Biopan	FluidPac	
Bion-8	1987				Interkosmos - IBMP
Bion-9	1989				Interkosmos - IBMP
Foton-7	1991				Glavkosmos ¹
Foton-8	1992		Х		TsSKB
Bion-10	1993	х			RKA - IBMP
Foton-9	1994		Х		RKA - Cosmos Group
Foton-10	1995	х			RKA - Cosmos Group
Foton-11	1997	х	Х		RKA - Cosmos Group
Foton-12	1999		Х	х	RKA - TsSKB ²
Foton-M1	2002		Х	Х	Rosaviakosmos ³ - TsSKB ²

- 1 Glavkosmos was a government authority set up in the last ten years of the USSR to market Soviet space capabilities abroad. Just before and after the demise of the Soviet Union, Glavkosmos was put forward as the contractual counterpart for flying foreign hardware on Russian spacecraft. The organisation ceased to exist after Foton-7 at least as far as ESA's contracting policy was concerned.
- 2 The name 'Cosmos Group' disappeared and was replaced by that of the Group's main member, TsSKB.
- 3 Following a reorganisation, RKA (the Russian Space Agency), changed its name in October 1999 to Rosaviakosmos (Russian Aviation and Space Agency)

decision to commit no more ESA experiments to the external exposure facilities (KNAs). Only one out of four KNAs on Bion-10 had been hermetically closed at the end of the flight (see Chapter 7). This led to contamination of the environment around the experiment packages and caused them to be subjected to excessive temperatures. Just three months earlier, the ESA Biopan facility had passed its in-flight qualification. With all the built-in design advantages over the KNAs, and its capability to fly on all types of spacecraft in the Foton class (i.e. Foton, Bion and Resurs-F), Biopan was ESA's logical successor to the KNA. Another decision was to cease flying radiobiological experiments inside the capsule, to avoid unwanted effects from the onboard radioactive caesium source (see Section 5.1.2).

Negotiations with the BION Company were suspended during the Bion-10 campaign, and were later resumed in the spring of 1993. However, in the meantime RKA had affirmed its position as the implementing organisation in the terms of the ESA-Russia "Cooperation Agreement" (see Chapter 1). When the BION offer was eventually submitted to ESA, RKA had designated the Cosmos Group as the responsible entity for Biobox and Biopan missions on unmanned Russian spacecraft.

At the end of March 1993, ESA and the consortium comprising RKA and the Cosmos Group reached in-principle agreement on a frame contract for the conduct of microgravity missions on Foton-type spacecraft. However, the unrest and instability in Russia did not cease in 1993, and cumbersome and lengthy Table 10-1. Bion and Foton missions with ESA participation. For a list of ESA life science experiments completed on these missions, see Annex I. review iterations followed each other, troubled by personnel and organisational changes on the Russian side of the signature page. Finally, on 20 December 1993, a Frame Agreement for the Launch, Operation and Retrieval of ESA Microgravity Multi-User Facilities and Experiments was signed by ESA on the one side and on the other side by RKA and the Cosmos Group. Concrete payload missions were to be defined in "Mission Orders" in the framework of and subordinate to the Frame Agreement. The slow maturation process of the Frame Agreement did not prevent the parties from signing the first Mission Order in July 1993, covering the Biopan-1 flight on the Foton-9 spacecraft. (The Biopan test flight in 1992 on Foton-8 was retroactively referred to as Biopan-0).

The signing of the Biopan-1 Mission Order signalled the end of ESA's negotiations with the BION company headed by IBMP. In November 1993 ESA informed the BION company that it would fly all future Biobox and Biopan missions in the context of the Frame Agreement with RKA and the Cosmos Group. This decision did not directly affect the scientific cooperation between individual investigators of IBMP and ESA-sponsored scientists, which indeed continued in a number of areas. However, the discontinuation of the relation with IBMP in its aspired role as a mission management organisation made it more difficult for ESA as a permanent guest on the IBMP premises with its Moslab facility. In fact, with no truly ESA-IBMP joint missions in the pipeline, the raison d'être of Moslab (see Chapter 3) became somewhat undermined.

The first ESA missions contracted to RKA-Cosmos Group were Biopan-1 on Foton-9 [3,4] and Biobox-2 on Foton-10 [5], both with some Russian scientific involvement, but without joint experiments. These were followed by Foton-11 (with a combined Biopan-Biobox payload) and Foton-12 [6, 7, 8, 9, 10], which saw the debut of ESA's 182 kg FluidPac fluid physics facility. Future reflights of FluidPac and Biopan in 2002 on a mission referred to as Foton-M1 were approved by ESA's Microgravity Programme Board in June 2000. ESA, Rosaviakosmos and TsSKB signed the Mission Order for Foton-M1 on 11 April 2001 at ESA HQ, Paris [11].

So much for the programmatic changes that occurred during the hectic final decade of the 20th century. ESA's Biopan and Biobox, developed in the early 1990s for Bion, are flying nowadays on Foton (Biopan) and the US Space Shuttle (Biobox). The two facilities can now be considered as 'veterans': both have completed four flights, with a fifth in the planning.

A list of the Bion-Foton missions with ESA participation is presented in Table 10-1. The rapid increase in the payload mass flown by ESA on Bion-Foton spacecraft is illustrated in Figure 10-1. In Annex I, the reader will find an overview of all ESA life science experiments conducted to this date on Bion and Foton (total number: 50).

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Annex I

ESA Life Sciences Experiments on Russian Recoverable Spacecraft 1987-1999

	Principal investigator(s)	Expt name	Experiment title	Proposal code	Facility	Organisms/samples
Bio	n-8 / Cosmos 1887 (1987)					
1	H. Bücker, A.M. Alpatov	CARAUCOS 1	Study of the effects of microgravity and HZE particles of cosmic radiation on the embryogenesis of the stick insect	BIOK-88-8-D	standalone	insect eggs
2	G. Reitz, V.E. Dudkin	DOSICOS 1	Radiation dosimetry inside and outside the spacecraft; radiation damage in plant seeds and bacterial spores	BIOK-88-9-D	KNA / standalone	bacterial spores
3	A.R. Kranz, V.V. Shevchenko	SEEDS 1	Biological effects of high and lower ionising cosmic heavy particles in genetically variable plant tissues	BIOK-88-11-D	KNA / standalone	plant seeds
Bio	n-9 / Cosmos 2044 (1989)					
4	H. Bücker, A.M. Alpatov	CARAUCOS 2	Study of the effects of microgravity and HZE particles of cosmic radiation on the embryogenesis of the stick insect	BIOK-88-8-D	standalone	insect eggs
5	G. Reitz, V.E. Dudkin	DOSICOS 2	Radiation dosimetry inside and outside the spacecraft; radiation damage in lettuce seeds	BIOK-88-9-D	KNA / standalone	plant seeds
6	R. Marco, J. Miquel, I.A. Ushakov	FLIES 1	Effects of microgravity and radiation on fruit fly development, ageing, rate of mitotic recombination and adaptation to the space environment	BIOK-88-14-E & BIOK-88-15-E	standalone	fruit flies
7	O. Rasmussen, F. Gmünder, M.G. Tairbekov	PROTODYN	The effect of the microgravity environment on the regeneration of plant protoplasts	BIOK-88-17-DK & BIOK-88-4-CH	standalone	plant protoplasts
8	A.R. Kranz, V.V. Shevchenko	SEEDS 2	Biological effects of high and lower ionising cosmic heavy particles in genetically variable embryonic plant tissues	BIOK-88-11-D	KNA / standalone	plant seeds

9	N. Chayen	KASHTAN	Protein crystallisation in microgravity: HIV reverse transcriptase	_	Kashtan	proteins
Fot	con-8 (1992)					
	Biopan test flight; no ESA-spons	ored scientific experim	nents		Biopan-0	
Bio	on-10 / Cosmos 2229 (1992/93)					
10	H. van den Ende, O.V. Gavrilova	ALGAE 1	Changes in the cell division cycle of Chlamydomonas monoica caused by microgravity	BIOK-88-6-NL	standalone	algae
11	J.P. Veldhuijzen, N.V. Rodionova	BONES	Growth and mineralisation of foetal mouse bones in microgravity	IMMU-88-8-NL	Biobox-1	embryonic mouse bones
12	I.A. Ushakov, R. Marco	CLOUD	The impact of pre-flight gravity stress on in-flight fitness in Drosophila melanogaster	-	standalone	fruit flies
13	M.G. Tairbekov	FIBRO 1	Morpho-physiological properties and differentiation of cell culture fibroblasts under conditions of spaceflight (microgravity)	-	Biobox-1	fibroblasts
14	C. Alexandre	OBLAST 1	Osteoblastic cells in weightlessness: morphology and biochemical response	BIOK-88-12-F	Biobox-1	osteoblasts
15	G. Schoeters, N.V. Rodionova	MARROW 1	Effect of microgravity on <i>in vitro</i> cultures of pre-osteoblast like cells	BIOK-88-16-B	Biobox-1	osteoblasts
16	J. Miquel, R. Marco & I.A. Ushakov	FLIES 2	Test of the metabolic hypothesis of accelerated ageing in space in Drosophila melanogaster	BIOK-88-14-E & BIOK-88-15-E	standalone	fruit flies
17	A.R. Kranz, JU. Schott, V.V. Shevchenko, A.M. Marenny	SEEDS 3	Analysis of chromosomal damage of marker lines in Arabidopsis seeds by cosmic heavy particles, including protons	BIOK-88-11-D	KNA / standalone	plant seeds
18	G. Reitz, A.I. Vikhrov	DOSICOS 3	Identification and quantification of incident radiation particles during orbital spaceflight	BIOK-88-9-D	KNA / standalone	none
19	L.V. Nevzgodina, G. Reitz	WOLFFIA	Radiation damage in Wolffia arrhiza caused by heavy ions of cosmic rays	-	standalone	duckweed

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Foton-7 (1991)

Foton-9 (1994)

20	J. Cadet	BASE 1	Base damage induced by cosmic radiation in cellular DNA	BP-91-4-F	Biopan-1	plant seeds
21	A. Hernandorena	SHRIMP 1	Radiation effects in gastrulae from the brine shrimp Artemia	BP-91-9-E	Biopan-1	brine shrimp embryos
22	J-P. Moatti	VITAMIN 1	Radiation effects and efficiency of radioprotective substances in biological acellular systems	BP-91-10-F	Biopan-1	lipoproteins
23	G. Reitz	MAPPING 1	Radiation measurements behind defined shielding	BP-91-11-D	Biopan-1	none
24a	G. Horneck	SURVIVAL 1	Effects of the harsh space environment on micro-organisms	BP-91-13-D	Biopan-1	bacterial spores; fungal spores
24b	G. Horneck	SHUTTER 1	Biological UV dosimetry	BP-91-13-D	Biopan-1	bacterial spores
25	A. Brack	DUST 1	Processing and stability of biomonomers in artificial dust grains	BP-91-14-F	Biopan-1	peptides
Fot	on-10 (1995)					
26	R. Bouillon	MARROW 2	Effects of microgravity in MG-63 human osteosarcoma cells	BIOK-88-16-B	Biobox-2	osteoblasts
27	C. Alexandre	OBLAST 2	Osteosarcoma cell culture from rat in weightless- ness: morphology and biochemical response	BIOK-88-12-F	Biobox-2	osteoblasts
28	M.G. Tairbekov, Ch. Lapiere	FIBRO 2	Morpho-physiological properties and differentiation of cell culture fibroblasts under conditions of spaceflight (microgravity)	-	Biobox-2	fibroblasts
29	R. Marco	FLIES 3	Test of the metabolic hypothesis of accelerated ageing in space in Drosophila melanogaster	BIOK-88-14-E	standalone	fruit flies
30	H. van den Ende	ALGAE 2	Changes in the cell division cycle of Chlamydomonas monoica caused by microgravity	BIOK-88-6-NL	standalone	unicellular green algae
31	W. Rietveld	BEETLE 1	Biological clocks of beetles: reactions of free-running circadian rhythms to microgravity	-	standalone	desert beetles

32	J. Cadet	BASE 2	Base damage induced by cosmic radiation in cellular DNA	BP-91-4-F	Biopan-2	plant seeds
33	A. Hernandorena	SHRIMP 2	Radiation effects in gastrulae from the brine shrimp Artemia	BP-91-9-E	Biopan-2	brine shrimp embryos
34	J-P. Moatti	VITAMIN 2	Radiation effects and efficiency of radioprotective substances in biological acellular systems	BP-91-10-F	Biopan-2	lipoproteins
35	G. Reitz	MAPPING 2	Radiation measurements behind defined shielding	BP-91-11-D	Biopan-2	none
36a	G. Horneck	SURVIVAL 2	Effects of the harsh space environment on micro-organisms	BP-91-13-D	Biopan-2	halophilic microbes; bacterial spores
36b	G. Horneck	SHUTTER 2	Biological UV dosimetry	BP-91-13-D	Biopan-2	bacterial spores
37	A. Brack	DUST 2	Processing and stability of biomonomers in artificial dust grains	BP-91-14-F	Biopan-2	peptides
38	R. Bouillon	MARROW 3	Effects of microgravity in MG-63 human osteosarcoma cells	BIOK-88-16-B	Biobox-3	osteoblasts
39	C. Alexandre	OBLAST 3	Osteosarcoma cell culture from rat in weightlessness: morphology and biochemical response	BIOK-88-12-F	Biobox-3	osteoblasts
40	M.G. Tairbekov, Ch. Lapiere	FIBRO 3	Morpho-physiological properties and differentiation of cell culture fibroblasts under conditions of spaceflight (microgravity)	-	Biobox-3	fibroblasts
41	R. Marco	FLIES 4	Test of the metabolic hypothesis of accelerated ageing in space in Drosophila melanogaster	BIOK-88-14-E	standalone	fruit flies
42	H. van den Ende	ALGAE 3	Changes in the cell division cycle of Chlamydomonas monoica caused by microgravity	BIOK-88-6-NL	standalone	unicellular green algae
43	W. Rietveld	BEETLE 2	Biological clocks of beetles: reactions of free-running circadian rhythms to microgravity	_	standalone	desert beetles

Foton-11 (1997)

Foton-12 (1999)

44	N. Dousset	VITAMIN 3	Radiation effects and efficiency of radioprotective substances in biological acellular systems	BIO-NSS-95-4-F	Biopan-3	lipoproteins; liposomes
45	G. Reitz	DOSIMAP	Dosimetric mapping	BIO-NSS-95-35-D	Biopan-3	none
46	G. Horneck	SURVIVAL 3	Effects of solar UV on micro-organisms	BP-91-13-D	Biopan-3	halophilic microbes; bacterial spores
47	J. Kiefer	YEAST	Radiation damage in yeast: interaction of space radiation components	BP-91-3-D	Biopan-3	yeast
48	A. Brack	STONE 1	Thermal processing of artificial sedimentary meteorites during atmospheric reentry	unsolicited	standalone	rocks
49	G. Briarty	SYMBIO	Plant-bacterial symbiosis in microgravity	BIO-NSS-95-21-UK	standalone	plant seedlings; nodule-forming bacteria
50	H. van den Ende	ALGAE 4	Effects of microgravity on cell cycle kinetics in the unicellular alga Chlamydomonas monoica	BIO-NSS-95-7-NL	standalone	unicellular green algae

Annex II ABBREVIATIONS and ACRONYMS

Throughout this document, the S.I. system of units and the corresponding internationally recognised abbreviations are used. In addition the following acronyms and abbreviations are used:

ACTA ALP	Academisch Centrum voor Tandheelkunde Amsterdam (NL) alkaline phosphatase
В	Belgium
BB	'Biologicheskij blok'; container for standalone experiments
BGJb	a cell culture medium, originally developed by Biggers, Gwatkin and Judah.
BSA	bovine serum albumin
BX	a file containing pre-processed telemetry data
Biobox	automated incubator for space biology developed by ESA
Biopan	exposure facility for space biology developed by ESA
Ci	curie
CIS	Commonwealth of Independent States
CIS	Cells in Space
CNES	Centre Nationale d'Etudes Spatiales (French Space Agency)
CPS	characters per second
CRN	Centre de Recherche Nucléaire (F)
CSDP	Control Spacialized Design Pursey (PUS)
CSDB	Ce Investigator
COL	
CN	cellulose nitrate
CND	cellulose nitrate manufactured by Daicel
CNK	cellulose nitrate manufactured by Kodak
CNR-CNUCE	Department of Space Flight Dynamics of the National Italian Research Centre in Pisa (Italy)
CR39	diallylglycol carbonate
CSLM	confocal scanning laser microscope
D	Germany (Deutschland)
D-1	Space Shuttle mission STS-61A (1985)
DLR	Deutsche Forschungsanstalt für Luft- und Raumfahrt (German Aerospace Centre)
DS	a file containing raw telemetry data
ECU	electrical control unit
EOF	eclipse orbit fraction
ESA	European Space Agency
ESOC	ESA's European Space Operations Centre at Darmstadt, Germany
ESTEC	ESA's European Space Research and Technology Centre at Noordwijk The Netherlands
eV	electron volt (a measure of the energy of a charged particle)
F	France
FC	Facility Controller
q	(in italic) the mean acceleration due to gravity on Earth (approx 9.81 m/s ²)
GMT	Greenwich Mean Time
Gy	Gray (1 Gy = 1 Joule per kg = 100 rad)
HZE	high-energy heavy ion
Ι	Italy
ICRP	International Commission for Radiological Protection
IBMP	Institute for Biomedical Problems (RUS)
IKI	Institut Kosmicheskikh Isslédovanii (Institute for Space Research) (RUS)
IMI _1	Snace Shuttle mission STS-42 (1992)
117117-1	Space Shame mission 515-42 (1772)
KB	'Konstrúktorskoye Byuro'; Design Bureau (RUS)
KNA	'Kontéjner naúchnoj apparatúry' (exposure facility for space radiation experiments)

L	(time of) launch
LED	light-emitting diode
LET	linear energy transfer
LST	local sidereal time
MEM	minimum essential (cell culture) medium
MLI	multi-layer insulation
Moslab	ESA's laboratory in Moscow specially constructed and maintained for the Bion-10 mission
MT	Moscow time (GMT + 3h)
NORAD	North American Aerospace Defence Command
NEWS	file giving forecast telemetry down-link times
NL	The Netherlands (Nederland)
NASA	National Aeronautics and Space Administration (USA)
PBS	phosphate-buffered saline
pH	minus the logarithm to base ten of the relative activity of hydrogen ions in a solution.
PI	Principal Investigator
PKG	Perry's Kettering Group
PSU	polysulphone
PTH	parathyroid hormone
R	return; (time of) landing
RAAN	right ascension of the ascending node
RAM	random access memory
RCSRS	Research Centre for Space Radiation Safety (RUS)
RKA	Rossíjkoe Kosmícheskoe Agéntsvo (Russian Space Agency)
RSSS	Radiation Safety Service for the Spaceflights (RUS)
RUS	Russian Federation
s/c SCB SRON SSR	spacecraft Switch and Control Box Stichting Ruimte Onderzoek Nederland (Space Research Organisation Netherlands)
TEM TLD TRW TsUP TsSKB	transmission electron microscope thermoluminescence detector TRW Space & Electronics Group, USA Tsentr Upravléniya Polëtami (Flight Control Centre) (RUS Tsentrálnoye Spetsialzírovanoye Konstrúktorskoye Byuro (RUS) = Central Specialised Design Bureau (CSDB)
UA	Ukraine
USSR	Union of Soviet Socialist Republics
USA	Unites States of America
UTC	Universal Coordinated Time
VITO	Vlaamse Instelling voor Technologisch Onderzoek (B)
Z	atomic number

Annex III The Russian Alphabet

Russian words and names are transcribed using the British Standards Institute (B.S.I.) transliteration, with graphical accents placed on stressed syllables in order to facilitate their correct reading.

Upper case	Lower case	B.S.I. Transliteration
А	а	а
Б	б	b
В	В	v
Г	Г	g
Л	Д	d
Ē	e	e
Ë	ë	ë
Ж	ж	zh
3	3	Z
И	И	i
Й	й	i
Κ	К	k
Л	л	1
Μ	М	m
Н	н	n
0	0	0
П	п	р
Р	р	r
С	с	\$
Т	Т	t
У	у	u
Φ	ф	f
Х	Х	kh
Ц	ц	ts
Ч	Ч	ch
Ш	ш	sh
Щ	щ	shch
Ъ	Ъ	55
Ы	ы	У
Ь	Ь	,
Э	Э	eh
Ю	ю	yu
R	Я	ya



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