



Environmental impacts of 5G

A literature review of
effects of radio-frequency
electromagnetic field
exposure of non-human
vertebrates, invertebrates
and plants

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A literature review of effects of radio-frequency electromagnetic field exposure of non-human vertebrates, invertebrates and plants

Telecommunication networks use radio-frequency electromagnetic fields to enable wireless communication. These networks have evolved over time, and have been launched in successive generations. The fifth generation of telecommunication networks will operate at frequencies that were not commonly used in previous generations, changing the exposure of wildlife to these waves. This report reviews the literature on the exposure of vertebrates, invertebrates and plants to radio-frequency electromagnetic fields in anticipation of this change.

The review shows that dielectric heating can occur at all considered frequencies (0.4-300 GHz) and for all studied organisms. Summarising and discussing the results of a series of studies of radio-frequency electromagnetic field exposure of wildlife, the review shows that several studies into the effects of radio-frequency electromagnetic field exposure on invertebrates and plants in the frequency bands considered demonstrate experimental shortcomings. Furthermore, the literature on invertebrate and plant exposure to radio-frequency electromagnetic fields above 6 GHz is very limited. More research is needed in this field.

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Executive summary

1. Rationale

Wireless telecommunication is a widespread technology that uses radio-frequency (RF) electromagnetic fields (EMFs) to send information between users. Wildlife can be exposed to these waves, which will partially penetrate biological tissues. These internal fields can have biological effects. Exposure to RF-EMFs and the interaction between the EMFs and organisms will depend on the frequency of the waves. Fifth generation wireless telecommunication networks (5G) will be operating partly at new frequencies that are not very commonly found in the environment. These anticipated changes warrant a review of the existing literature on the effects of RF-EMF exposure of wildlife. This study presents such a review.

2. Methodology

A database search of the current literature in the field found that it is subdivided based on two classifiers. The first is the target group investigated: non-human vertebrates, invertebrates and plants; the second is the RF-EMF frequency studied, which is subdivided between a lower (0.45-6 GHz) and a higher frequency range (6-300 GHz). The former frequency range includes those frequencies where the current telecommunication networks operate, while the latter is the range in which 5G will partially operate. This resulted in six categories, which are reviewed separately.

3. Results

Dielectric heating due to RF-EMF exposure of biological tissue is shown in all categories. This heating causes internal temperature increases in organisms or cells, which in turn has biological effects such as a thermoregulatory response. This implies that there is always a level of RF-EMF power density that will cause biological effects, referred to as thermal effects. Decoupling effects caused by elevated temperatures and the presence of RF-EMFs within biological tissue are major issues in this field of study.

Many studies focus on demonstrating (the absence of) non-thermal effects. These are effects that are caused by RF-EMF exposure but are not associated with any changes in temperature. A wide variety of other effects of RF-EMF exposure are studied. However, no effect, apart from dielectric heating, is studied in all six categories.

Lower frequency range (0.45-6 GHz)

Vertebrates

In the lower frequency range, in vitro studies on non-human vertebrate cells showed mixed results on cellular genotoxicity and cellular transformation under RF-EMF exposure. Previous reviews on these subjects conclude either that the evidence for such effects is weak or that the literature is inconclusive. Regarding non-genotoxic effects of RF-EMF exposure, there are reports claiming that neural activity can be altered in vitro through RF-EMF exposure. Other cellular effects are either not proven or contested, or there are not enough studies to come to any conclusions on such effects. In vivo studies on genotoxicity of RF-EMFs found contradictory results. There is a debate in the literature on whether RF-EMF exposure can induce (transient) changes in the permeability of the blood-brain barrier.

It seems that the most recent studies could not show such effects. There are mixed results regarding the in vivo effects of RF-EMF exposure on the neural system. There seems to be a consensus that animals can hear (pulsed) RF-EMFs above a certain threshold, so-called microwave hearing. However, there is little evidence that telecommunication signals can induce this effect. Environmental studies on RF-EMF exposure and vertebrate behaviour focus mainly on animal

nesting, reproduction, orientation and abundance near RF-EMF sources. There are a limited number of studies that conclude that behavioural and reproductive effects might occur for birds and bats under RF-EMF exposure.

Invertebrates

RF-EMF exposure of invertebrates in the lower frequency range has been studied by several authors. In addition to dielectric heating, there is a focus on developmental, genetic, or behavioural effects. In vitro studies have shown increased neural activity in invertebrate neurons. In vivo studies on invertebrates are faced with several experimental problems and present inconclusive results on a series of investigated parameters. More research of higher quality, sham-exposed control groups is necessary. As for the limited number of studies that investigated non-insect invertebrates, they all found effects (in vitro and in vivo). This calls for more research on this topic. A very limited number of environmental studies focus on invertebrates and studies on non-insect invertebrates are under-represented as well. These topics require more research in the future.

Plants and fungi

Dielectric heating of plants has been shown in the lower frequency range. This heating might have beneficial effects, but will also induce plant mortality at a certain level. At lower levels of RF-EMF exposure, the literature on plants and fungi shows contradictory results and is plagued by experimental shortcomings. The numbers of studies and plants studied, especially for fungi, is limited in comparison to those studies that focus on animals. More research in this area is necessary, and should focus on a higher quality of unexposed control and sham control groups, temperature and exposure monitoring, and dosimetry.

Higher frequency range (6 to 300 GHz)

Vertebrates

In the higher frequency range, in vitro studies on both vertebrate and invertebrate neurons have shown effects of RF-EMF exposure on neural activity. In vivo studies on vertebrates have shown that RF-EMF exposure of the eye can induce corneal lesions and cataract. Effects on male fertility have been demonstrated in rodents as well. Mixed results of RF-EMF exposure on behaviour and prevalence of vertebrates are found. One research group demonstrated that RF-EMF exposure can have a hypoalgesic effect in mice. These effects should be replicated by other research groups. There is some evidence that high-frequency RF-EMFs can be used to induce an anti-inflammatory response, up to a certain dosage. A limited number of in vivo studies have shown that high-frequency RF-EMFs can reduce tumor growth.

Invertebrates

In the same frequency range, there have been in vitro demonstrations of neurostimulation and in vivo demonstration of developmental and teratogenic effects on invertebrates at relatively high power-densities. These effects should be investigated further at lower power densities. The literature on invertebrate exposure to RF-EMFs in this frequency range is limited and warrants further investigation.

Plants and fungi

The literature on fungi and plants in the higher frequency range is very limited and no conclusions besides the existence of dielectric heating can be drawn at this moment. It is necessary to execute further research in this area.

4. Conclusions

Dielectric heating due to RF-EMF exposure is shown in all categories studied.

In the lower frequency range (0.45-6 GHz), the majority of the existing literature focuses on vertebrates, for which a series of potential effects are studied. Those studies that investigate RF-EMF exposure of invertebrates in the lower frequency range focus on dielectric heating, and developmental, genetic or behavioural effects. Literature on non-insect invertebrates is very limited. Studies on plant exposure in the lower frequency range, which target exposure outcomes at plant level show experimental shortcomings. The number of studies in this category is limited in comparison to those studies that focus on animals.

In the higher frequency range (6-300 GHz) the number of peer-reviewed publications is in general lower than in the lower frequency range. For vertebrates, a series of potential exposure outcomes are studied, while the literature on invertebrates and plants above 6 GHz is very limited. More research in this field is necessary.

5. Policy options

Given the results of this review, four policy options were formulated.

A first policy option could be to fund research on RF-EMF exposure of plants, fungi and invertebrates at frequencies below 6 GHz and to fund research on non-human vertebrates, plants, fungi and invertebrates at frequencies of between 6 and 300 GHz. These studies could form the basis for evidence-based policies regarding RF-EMF exposure of non-human organisms.

A second policy option could be to call for systematic monitoring of environmental RF-EMFs, since these are the main source of exposure for non-human organisms and it is expected that this exposure will change over time.

A third policy option could be a request to make information on the RF-EMF operational aspects of the telecommunication networks public. This would again be aimed at quantifying environmental RF-EMF exposure over time.

A fourth policy option could be to require compliance studies for organisms other than humans when base station antennas are installed in the telecommunication network. This would prevent the excessive RF-EMF exposure of non-human organisms near such antennas.

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1. Introduction

1.1. Wireless telecommunication systems

Wireless telecommunication is a widespread and growing technology in Europe. This wireless telecommunication is in most cases enabled by Electromagnetic fields (EMFs). These are commonly characterized by their wavelength or frequency. A frequency expresses the number of oscillations of a wave per unit of time in Hz or s^{-1} . EMFs with higher frequency have higher energy and are able to ionize molecules and atoms. Therefore, this frequency range of EMFs is called ionizing radiation. The EMFs that are used in wireless telecommunication systems are located at lower frequencies and cannot ionize matter. These are consequently classified as non-ionizing radiation ([ICNIRP_2020](#)). Radio-frequency (RF) EMFs are a particular range of non-ionizing waves, located at frequencies from several kHz up to 300 GHz.

Wireless telecommunication connects different users of a wireless network. In most large-scale telecommunication networks these users are not connected directly to one another, but are connected to one or more intermediate network providers. These telecommunication providers operate a wireless network connected to a wired backbone network. This wireless network covers the areas in which users can request service. In order of increasing area these are described as: atto-, femto-, pico-, micro-, and macro-cells. The users are served by the networks through the emission of RF EMFs that carry the signals. These RF EMFs are emitted and received by antennas.

Antennas are the intermediary structure in between guided and freely propagating EMFs. The antennas that make up the providers' wireless networks are diverse. The largest variety are base station antennas (BSAs) (Thielens et al. 2013). These are antenna arrays (a collection of collaborating antennas arranged in a pattern), which are typically mounted on the side or top of towers or tall buildings. BSAs are used in macro-cells, which are areas of several square kilometers in which a relatively large number of users is covered. In order to cover such large areas these BSAs are fed hundreds of Watts of power. On the other side of the spectrum of antennas available to network providers, one can find atto- and femto-cell antennas, which are smaller antennas, sometimes integrated in existing structures (Torfs et al. 2018), that cover areas of several square decimeters to square meters. These types of antennas emit lower RF power and are deployed in offices or residential areas. The users are connected with the network through their personal, mobile devices. These devices contain miniaturized antennas that are often customized in design for a particular user device (Rowell and Lam 2012).

Telecommunication relies on bidirectional wireless traffic between the network on the one hand and the users on the other hand or in some cases directly between two users. The wireless link in which information is sent from the network towards to user is referred to as downlink (DL), while the opposite direction is denoted uplink (UL). Some wireless technologies are unidirectional. In this case, the users only receive a signal from the network and do not send any information towards to network. This is referred to as broadcasting. Typical examples are wireless radio- and television broadcasts.

The RF frequency spectrum is regulated and there are particular frequency bands in which telecommunication is allowed. In the European Union (EU), the Electronic Communications Committee (ECC) within the European Conference of Postal and Telecommunications Administrations (CEPT) is responsible for the (future) planning and harmonization of the RF spectrum in the European Union (EU) (<https://efis.cept.org/>). There are small differences between the different EU member states (ECC 2019). The current networks rely on frequencies between 0.1 GHz and 6 GHz (Bhatt et al. 2016). In the EU, the frequency bands that are most commonly studied

with relation to personal exposure to RF-EMFs (Velghe et al. 2019a) are listed in Table 1. A full overview of all the allocated frequency bands in the EU can be found in (ECC 2019).

Table 1 – Overview of the most commonly studied telecommunication frequency bands (Velghe et al. 2019a)

System	Frequency (MHz)
FM Radio	87.5-108
DVB	470-790
800 DL	791-821
800 UL	832-862
900 UL	880-915
900 DL	925-960
1800 UL	1710-1785
1800 DL	1805-1880
DECT	1880-1900
2100 UL	1920-1980
2100 DL	2110-2170
WLAN	2400-2485
2600 UL	2500-2570
2600 DL	2620-2690
WLAN	5150-5875

Table 1 lists several frequency bands in which are assigned to a wireless technology. Frequency Modulated (FM) Radio is a broadcasting technology that is used for radio transmissions around. Digital Video Broadcasting (DVB) is a broadcasting technology that is used to transmit digital television emissions towards the users. Digital Enhanced Cordless Telecommunication (DECT) is a telecommunication technology that is used for communication between a small base station and (multiple) cordless phones. Wireless Local Area Network (WLANs) are small to medium sized networks that are used for wireless internet access at home or in a professional context. A Wireless Fidelity (Wi-Fi) network is an example of a WLAN. Those frequency bands in Table 1 that are named using a frequency and UL or DL, used to be assigned to a certain telecommunication technology.

However, in recent years these technologies have been redistributed and spread over the different frequency bands instead of being allocated to a fixed frequency band. The most common technologies are Global System for Mobile communications (GSM), Universal Mobile Telecommunications System (UMTS), and Long-Term Evolution (LTE), which were the telecommunication technologies that were launched in the 2nd, 3rd, and 4th generation of telecommunication networks, respectively.

1.2. Exposure to wireless telecommunication systems

Since wireless telecommunication systems are so widespread, many animals and plants are exposed to RF-EMFs. There exists a variety in RF-EMF exposure scenarios. The type of scenario is classified according to the source parameters and the exposed organism. In general, the source can either be internal to the organism (for example an implant), in direct contact with the organism (for example

high-frequency electrodes), or the source can be external to the organism (for example a base station antenna). Depending on the type and configuration of the source and the RF-EMF frequency the exposure can be a whole-body exposure, i.e. an exposure scenario in which the whole organism is (uniformly) exposed to RF-EMFs, or a localized exposure, i.e. an exposure in which only a part of the organism receives a significant amount of RF-EMFs. For an external RF-EMF source, the exposure scenario is divided in several categories, depending on the separation distance between the source and the organism. In the far-field, the distance between the RF-EMF source and the exposed organism is $2D^2/\lambda$, where D is the maximal dimension of the source or the organism and λ is the wavelength. When the source is closer to the organism, this is often described as near-field exposure. Often, far-field sources cause whole-body exposures, while near-field sources cause localized exposures. However, this is not true in all scenarios and is expected to change in future wireless networks (see Section 1.3).

These RF-EMFs can penetrate biological media and can be absorbed in such media (ICNIRP 2020). This absorption can be quantified using the specific absorption rate (SAR in W/kg), which is the amount of power absorbed in a certain mass. This quantity is only meaningful when averaged over a certain volume or mass. The whole-body averaged SAR is a commonly used quantity to estimate exposure to RF-EMFs in which the entire organism is exposed to RF-EMFs. This quantity is not always useful in a localized exposure scenario. Therefore, a smaller averaging volume or mass is required to characterize localized exposure. Such a volume or mass is then often defined in such a way that a threshold value of SAR averaged over that volume or mass corresponds to a biological effect. The field of science that investigates SAR under different exposure conditions is called RF-EMF dosimetry. There are other quantities that could be used to quantify RF-EMF exposure, if absorption of RF-EMFs is not of interest, magnitudes of internal electric and magnetic fields and magnitude of currents in biological tissue can be determined as well.

Often, it is not possible to measure and/or quantify the EMFs inside of an organism. Therefore, RF-EMF exposure is often quantified by studying the incident RF-EMF fields. These are the EMF fields that would be present on the location of an organism, if that organism would not be there. These incident fields induce the internal EMF fields (and absorption of these fields). This exposure can be quantified using the electric field strength (E in V/m), which is the amplitude of the electric field (E). Alternatively, RF-EMF exposure can also be quantified using the electromagnetic power density (S in W/m^2).

In free-space, i.e. without any interference or blocking caused by objects in the environment, both E and S decrease as function of distance from an emitting antenna (propagation losses). This is another important difference between near- and far-field exposure. The SAR induced by and the power density S around an antenna scale linearly with the RF input power in the antenna. The amplitude of the electric field strength scales quadratically with the input power.

In the case of an internal source, an RF-EMF source in direct contact with an organism, or near-field exposure to an external RF-EMF source, there is no fixed relationship between the RF-EMF magnitudes, the power density, and the SAR or internal field magnitudes. These exposure quantities have to be evaluated on a case-by-case basis. However, it is often possible to provide lower and upper bounds of the exposure. In case of an external source in the far-field of an organism there exists a fixed relationship between both power density and electric field strength ($S=E^2/377$).

In the literature on RF-EMF exposure of the general population, a differentiation is made between users and non-users of telecommunication networks. Both categories are exposed to environmental RF-EMFs that are emitted by the telecommunication networks and other users in the environment. These sources are often in the far-field of the exposed subject. However, users are also exposed to RF-EMFs emitted by their own devices in the near-field of the subject.

1.3. New aspects of fifth generation wireless telecommunication systems

1.3.1. Frequencies

The goal of fifth generation (5G) mobile networks is to enable significantly faster mobile broadband speeds and increased data usage. One of the technological changes that should enable these goals is the use of additional (higher) frequency bands in the RF-EMF spectrum. 5G pioneer bands identified at EU level are the 700 MHz (694 - 790 MHz), the 3.6 GHz (3.4-3.8 GHz) and the 26 GHz (24.25-27.5 GHz) frequency bands (Pujol et al. 2020).

1.3.2. Adaptive downlink transmissions

In the current networks DL transmission occurs using a fixed, wide beam that covers a sector of a cell. One of the goals of 5G networks is to serve multiple users simultaneously at the same carrier frequency using the same base station antenna. This requires an improvement in the signal-to-noise ratio (SNR) and signal-to-interference ratio (SIR) at each user. In order to increase SNR using a fixed beam, the total input power of the beam would have to go up. This is unwanted and does not provide a solution for SIR. Therefore, new ways of performing DL transmissions are used in 5G networks. One of the main approaches that will be used to achieve this is the use of adaptive transmissions from the antenna arrays in the base stations for DL data transmission towards the users (Marzetta 2010). In its most straightforward form, this approach tunes the phase and amplitude on each element of the antenna array in order to achieve a maximal received signal strength on the user's device (SNR optimization). As the user moves around in the network, these phases and amplitudes are adapted in order to keep a high SNR. In more complex forms, the phases and amplitudes on the base station elements are chosen in such a way that the fields at the intended user are elevated, while simultaneously reducing those fields at other users (SIR and SNR optimization) (Marzetta 2010). When a user is in line-of-sight (LOS) of a base station, such array precoding schemes result in the formation of a narrow beam towards the user (Thors et al. 2017). When a user is in obstructed LOS or non-line-of-sight (NLOS), then this results in a volume of elevated field strength around the user device (Shikhantsov et al. 2020).

1.3.3. Channel access methods

It is also expected that 5G networks will use new channel access methods. In the first roll-out of 5G, so-called 5G new radio (NR) the channel access method of choice is Time Division Duplexing (TDD) (Baracca et al. 2018; Thors et al. 2017). This method assigns the same frequency (blocks) to the UL and DL from the same user (or set of users). UL and DL are then assigned different times in which they can take place. Instead of assigning the full available bandwidth to all users, 5G NR will also assign part of the available bandwidth to specific users (Aerts et al. 2019). This allows for more options in the configuration of the network.

1.4. Exposure of wildlife to RF EMFs

The vast majority of wildlife, non-human vertebrates, invertebrates, or plants are not using a wireless technology or network. Therefore, in terms of RF-EMF exposure they will all fall into the non-user category. In this category the dominant sources of RF-EMF exposure are far-field sources, see Section 1.5. When comparing the exposure of plants to RF-EMFs with animals, the obvious difference is that plants are immobile and hence their orientation with relation to the RF-EMF base station antennas that make up the network is constant. Plants rely on high-frequency EMFs to perform photosynthesis and many have relatively high surface area to volume ratios in order to maximize exposure to sunlight. Obviously, this also makes them efficient receptors of other far-field EMF sources, such as most RF-EMF sources (Alain Vian et al. 2007). Temporal variation in RF-EMF exposure

of plants can occur due to temporal changes in the network and mobile users of RF-EMF that might appear in the vicinity of a plant while emitting RF-EMFs. The mobility of animals will induce more temporal variation in their RF-EMF exposure, since RF-EMF exposure of non-users has a spatial dependency (see Section 1.5).

While most non-human vertebrates will experience a small contribution of near-field exposure, the number of wireless technologies that generate near-field RF-EMF exposure of non-human vertebrates are increasing. Radio-tracking or radio-telemetry is a commonly used technique for monitoring vertebrates in the wild (White and Garrott 2012; Godfrey 2003; Millsbaugh and Marzluff 2001). Dedicated wireless networks have been deployed to perform RF-enabled tracking of animals in the wild (Panicker, Azman, and Kashyap 2019). There is also a growing number of wireless technologies in agriculture (S. Benaissa et al. 2017; Dlodlo and Kalezhi 2015; Said Benaissa et al. 2016).

There are some wireless applications that generate near-field RF-EMF exposure of invertebrates. Entomological radar is a technology that makes use of the scattering of EMFs by insects to detect them. In this radar approach, a pulse of RF-EMFs is emitted from the radar station towards an insect. The EMFs are then partially reflected from the insect and these reflected fields are received by the radar station. Entomological radar is used to study insect behaviour and proliferation (Chapman, Drake, and Reynolds 2011; Glover et al. 1966; Riley 1985). Wireless sensor networks targeted at monitoring insect pollinators exist (Edwards-Murphy et al. 2016; Henry et al. 2019; Kridi, de Carvalho, and Gomes 2016). There are also some telemetry studies for insects (Daniel Kissling, Pattemore, and Hagen 2014). This is a field in which an insect is tracked wirelessly by attaching an RF-tag to the animal, which sends information to a remote reader. Finally, RF-EMFs are used in agriculture for treatment of stored grains, nuts, and fruits (Das, Kumar, and Shah 2013a). It is expected that the application of these technologies will grow in the future.

The number of wireless monitoring tools in agriculture is increasing (Ruiz-Garcia et al. 2009). Wireless sensors networks are deployed in agriculture to monitor leaf growth (Palazzari et al. 2015; S. N. Daskalakis et al. 2018; Zhao et al. 2019), plant development (Burrell, Brooke, and Beckwith 2004), soil moisture (S.-N. Daskalakis et al. 2016; Vellidis et al. 2008), and other applications (Ruiz-Garcia et al. 2009). These applications now mainly occur using wireless technologies in license-free frequency bands such as Bluetooth and Zigbee (Ruiz-Garcia et al. 2009) and will generate near-field RF-EMF exposure of plants. It is conceivable that wireless solutions compatible with the 5G network will be rolled out in the future, also exposing plants to user-induced RF-EMF exposure.

1.5. Exposure of non-users in wireless telecommunication networks

Most of the organisms that fall into the studied categories in this review are not users of wireless telecommunication networks. Therefore, this section presents an introduction to the aspects of non-user exposure to RF-EMFs.

1.5.1. Current networks

Multiple methods for measuring the exposure of non-users of wireless telecommunication networks have been proposed. One approach uses so-called in situ measurements, where RF-EMF exposure is measured using a static receiving antenna, usually able to measure three orthogonal components of an incident EMF. Such an antenna is then combined with a spectrum analyzer that registers the received power as function of frequency (Joseph et al. 2009; Aerts et al. 2019). This method has a relatively low measurement uncertainty, but is time-consuming, requires a trained operator, and is stationary. This makes the method not suitable for population surveys of measurements that cover larger areas. Handheld and wearable devices are typically used when RF-EMF exposure of non-users

is measured. The commonly used measurement device for such measurements are personal exposimeters (Thielens, Van den Bossche, et al. 2018; Bolte 2016), which are body-worn devices that measure RF-EMF exposure in a set of frequency bands, see Table 1. (Röösli et al. 2010) proposed a protocol for the use of these measurement devices in studies that investigate the RF-EMF exposure of the population. (Neubauer et al. 2010; Bolte 2016) studied how the measurements that are done using personal exposimeters correspond to the actual RF-EMF exposure of non-users (and users).

These measurement methods have been used to investigate those factors that influence non-user RF-EMF exposure. There are spatial variations in non-user RF-EMF exposure (Bhatt et al. 2016; Bolte and Eikelboom 2012; P. Frei et al. 2009; Sagar et al. 2016; 2018; Thielens, Van den Bossche, et al. 2018; Urbinello, Huss, et al. 2014; Velghe et al. 2019b). These have been validated using EM geospatial simulations that take into account antenna parameters and EM propagation models (Beekhuizen et al. 2013; 2014; Bürgi et al. 2010). Population density has been put forward as a predictor of higher far-field RF-EMF exposure (Bhatt et al. 2016; Sagar et al. 2018; Velghe et al. 2019b). It has been shown by several studies that there are temporal variations of this non-user RF-EMF exposure (Aerts et al. 2018; Velghe et al. 2019b; Joseph et al. 2009; Bolte and Eikelboom 2012; Birks et al. 2018; P. Frei et al. 2009). Environmental variations and changes in wireless data traffic in the network are causing these temporal fluctuations (Joseph et al. 2009; Mahfouz et al. 2012). It was also investigated whether regulation on RF-EMF exposure has an influence on this non-user exposure. (Urbinello, Joseph, et al. 2014) performed measurements in three European countries with different regulations on RF-EMF exposure and found no significant effects of regulations. (Velghe et al. 2019b) investigated the effect of differences in regulation within Belgium and only found an effect on DL exposure, not on total exposure.

1.5.2. Fifth generation networks

The expectation in literature is that 5G will induce changes in RF-EMF exposure (Aerts et al. 2019). In the current wireless mobile technologies data transmission happens over fixed, cell-wide beams. This implies that DL exposure might change within a cell due to obstructions (buildings, vehicles, etc.) that might change the RF-EMF propagation towards a user. However, the base stations will perform no adaptations to their radiation pattern that are specific to a certain user or location. One of the main differences with the current networks is that exposure to base stations (DL exposure) will depend on whether an individual is a user of the network or not (Velghe et al. 2020).

DL exposure in a 5G network will be divided in three components (Velghe et al. 2020). First, there will be a broadcasting DL component, because the networks will send out a control signal from their base stations to find potential users within the network. Second, there will be an auto-induced data transmission DL component. This is a targeted transmission done from the base station towards the user with the goal of achieving data transmission. This can be in the form of a narrow beam aimed from the base station to the user's device or a zone of elevated RF-EMF strength at the user's device created using constructive interference. The third component will be induced by environmental data transmission (traffic) DL signals. This is exposure that is generated by targeted transmissions aimed from base stations at other, nearby users. A non-user will not experience the auto-induced component of DL exposure, which is expected to be the dominant component of the DL exposure (Baracca et al. 2018). Besides DL exposure a non-user will also be exposed to UL signals from nearby users. The current telecommunication networks mainly use RF-EMF frequencies below 6 GHz (Bhatt et al. 2016). However, new frequencies higher than 6 GHz will be used in 5G networks (Pi and Khan 2011; Pujol et al. 2020). It has already been demonstrated that internal EMFs (Bakker et al. 2011; Thielens et al. 2013) depend on the frequency of the incident EMFs. Hence, also for non-user exposure there will be a change in exposure as the use of frequencies in telecommunication systems shift.

Besides these changes in the physical layer, there will also be new channel access methods (Thors et al. 2017) and network architectures (Torfs et al. 2018). All these factors will alter non-users'

exposure in 5G networks. This exposure can be quantified using in-situ field measurements (Aerts et al. 2019), once 5G networks are deployed in the environment.

1.6. Rationale and goals of review study

The previous sections of this introduction have shown that wildlife will be exposed to RF-EMF and that this exposure will change in 5G networks. The existing review studies on effects of RF-EMF exposure of wildlife have been executed without knowledge of these developments. Hence, it is plausible that the current knowledge of these anticipated changes might lead to a reinterpretation of the existing literature on effects of RF-EMF exposure of wildlife. In particular the anticipated change in telecommunication frequency is a factor that has not been the focus of any previous review study on RF-EMF effects on wildlife.

Therefore, the goal of this study is to review literature that focuses on effects on wildlife (flora and fauna) due to exposure to RF-EMFs both at the current and future frequencies that will be used for telecommunication. To this aim, a database search of current literature in this field will be executed. This literature will be subdivided into two categories: first studies evaluating effects due to exposure to RF EMFs at lower frequency range (450 to 6000 MHz). This range also includes frequencies used in previous generations broadband cellular network. The second category will include studies that investigated exposure to RF EMFs at higher frequency range (6 to 300 GHz). Both subsets will be summarized and reviewed and conclusions will be drawn on those effects that have been shown in literature.

2. Methodology

This section presents the methodology used in reviewing existing literature on effects of exposure of wildlife (flora and fauna) to RF EMFs in 5G networks.

2.1. Population

All studies that were obtained from the literature search are divided in three taxonomy groups: (1) invertebrates, (2) vertebrates, and (3) plants and fungi. All human studies were excluded from the vertebrate's category. Studies that investigated RF-EMF exposure of vertebrates, invertebrates, and plants population both in vivo and in vitro were included. Additionally, observational studies, population studies, and exposure assessment studies that target the previously mentioned taxonomy groups.

2.2. Exposure

Studies are included that evaluated the exposure to RF EMFs used in telecommunication networks, choosing in particular the frequencies that were established as standard for use from the European Union: 450 MHz - 300 GHz. These are divided in two categories:

- Currently used telecommunication frequencies: 450 MHz – 6 GHz
- Newly used telecommunication frequencies: 6 GHz – 300 GHz

The first category is further referred to as 'Low Frequencies', while the second one is further referred to as 'High Frequencies'. In combination with the three studied taxonomies, this resulted in six categories that were defined prior to the analysis of the available literature:

- 'Low frequencies' (450 MHz - 6 GHz): evidence of effects on vertebrates
- 'Low frequencies' (450 MHz - 6 GHz): evidence of effects on invertebrates
- 'Low frequencies' (450 MHz - 6 GHz): evidence of effects on plants and fungi
- 'High frequencies' (6 - 300 GHz): evidence of effects on vertebrates
- 'High frequencies' (6 - 300 GHz): evidence of effects on invertebrates
- 'High frequencies' (6 - 30 GHz): evidence of effects on plants and fungi

Further subdivision in subcategories was done after the database search was analyzed, see Section 2.4.

2.3. Outcomes

Studies that investigated the following effects that have been associated with RF-EMF exposure were included: reproductive effects, morphogenesis, carcinogenicity, hyperthermia, dielectric heating, cataract, development, orientation, movement mechanisms, population diversity and abundance, behavioural effects, magnetic sense, neural effects, genotoxicity, gene expression, protein expression, cardiovascular effects, auditory effects, cerebral effects, and physiological effects. Studies that include untreated or sham-treated populations (controls and sham-controls) are preferred, but other study designs are included in the review as well.

2.4. Database search

Studies are not selected based on the study design. However, non-original studies were excluded. The review was initially restricted to peer-reviewed journal articles published from 1945 on, in English. The presence of the journal in the ISI Web of science was used as criterion for being peer-

reviewed. However, other publications used as references in the resulting set of publications were also included in the dataset, in case they were in English and focused on one of the studied populations and one of the studies exposures and one of the studied outcomes.

A systematic search of the Web of Science following electronic academic databases was executed for potentially eligible records. The following keywords were used to build search strings suitable for the databases:

EXPOSURE: EMF; EMR; RFR; 5G; fifth generation; radiofrequency radiation; radiofrequency; radio-frequency; electromagnetic; electromagnetic field; electromagnetic radiation; millimeter wave; microwave

and

POPULATION (wildlife): wildlife; biodiversity; fauna; animal(s);

or

POPULATION (vertebrates): in vivo; rodent(s); rat(s); mouse; mice; vertebrate(s); mammal(s); fish; amphibian(s); bird(s).

or

POPULATION (invertebrates): invertebrate(s); insect(s); arthropod(s); mollusks: Mollusca; annelids; worm(s); snail(s); Cnidaria; Cnidarian(s); Arachnid(s); Arachnida; Crustaceans; Crustacea; Coral(s); Anthozoa; Echinoderm(s); sponge(s); jellyfish

or

POPULATION (plants): plant(s); tree(s); flower(s); plantae; algae; fungi; moss(es); fern(s);

The resulted literature was screened for relevance and relevant information was obtained from the paper to synthesize the evidence from selected literature. All the results clearly outside the RF-EMF field of research and those that clearly did not investigate an effect of RF-EMFs were excluded. Specific inclusion and exclusion criteria and subcategorizations were then applied per category.

2.5. Post-processing

In all categories, except 'Low-Frequency Vertebrates', the retained papers were subdivided in two types: one type of studies were only listed by not summarized and discussed (Type I) and the other type of studies are summarized, discussed, and tabulated (Type II). The type of outcome and subjects was different for each category, as different effects of RF-EMF exposure are studied in different categories. A grouping into outcomes of subjects is performed separately in each category.

2.5.1. Low frequency – vertebrates

In this category a meta-analysis of review studies that investigated RF-EMF exposure of vertebrates in the 0.45-6 GHz frequency range was executed. This means that original research papers (in vivo, in vitro, experimental, observational, population, exposure assessment, dosimetry, and studies focused on dielectric parameters) were not reviewed and are also not listed in this category. Only peer-reviewed papers or reports that were referred to in peer-reviewed paper that reviewed a series of or one effect caused by RF-EMF exposure on non-human vertebrates or vertebrate cells were included. The conclusions of the different review papers are compared and synthesized. In this category, exposure outcomes in lab animals related to cancer, reproduction, and development were not included in this study, because they are reviewed in a parallel study ordered by the STOA (Effects on health of fifth generation wireless communication).

2.5.2. Low frequency – invertebrates

In the category of studies on RF-EMF exposure of invertebrates in the 0.45-6 GHz frequency range, the following type of study subjects were classified as Type I: reviews, dosimetry, wireless monitoring, and dielectric properties. The following type of study subjects were included as Type II: dielectric heating, experimental and observational studies on insects, and experimental studies on other invertebrates.

2.5.3. Low frequency – plants and fungi

In the category of studies on RF-EMF exposure of plants and fungi in the 0.45-6 GHz frequency range, the following type of study subjects were classified as Type I: reviews, dosimetry, cellular and molecular studies, and dielectric properties. The following type of study subjects were included as Type II: dielectric heating, experimental and environmental studies.

2.5.4. High frequency – vertebrates

In the category of studies on RF-EMF exposure of vertebrates in the 6-300 GHz frequency range, the following type of study subjects were classified as Type I: reviews, dosimetry, dielectric properties, and studies that did not fit into a group of study outcomes (other studies). The following type of study subjects were included as Type II: cellular studies and animal studies.

2.5.5. High frequency – invertebrates

In the category of studies on RF-EMF exposure of invertebrates in the 6-300 GHz frequency range, the following type of study subjects were classified as Type I: reviews, dielectric properties, and studies that did not fit into a group of study outcomes (other studies). The following type of study subjects were included as Type II: dielectric heating, experimental studies on insects and spiders, and studies on neural activity.

2.5.6. High frequency – plants and fungi

In the category of studies on RF-EMF exposure of plants and fungi in the 6-300 GHz frequency range, the following type of study subjects were classified as Type I: reviews, dielectric properties, imaging, and remote sensing. The following type of study subjects were included as Type II: single-celled fungi and multi-cellular plants.

3. Results

Results in this section are presented according to the categories defined in Section 2.2, i.e. a division in frequency ranges and taxonomy group.

3.1. Lower telecommunication frequencies (450 MHz - 6 GHz)

3.1.1. Review of effects on vertebrates

Overview

This section is the result of a meta-analysis of 45 prior reviews that investigated RF-EMF exposure of non-human vertebrates, see Table 2. The studies in this field can be subdivided in three groups: in vitro studies or cellular studies that investigate effects and exposure on a cellular level in an experimental/laboratory context, in vivo or animal studies that investigate the exposure of the animal as a whole in an experimental/laboratory context, and environmental studies where the exposure is not generated experimentally, but present in the environment. In the cellular studies, the considered outcomes are genotoxicity, cellular transformation, and non-genotoxic cellular effects. In the animal studies, the investigated outcomes are: genotoxicity, carcinogenic effects, reproduction and development, effects on the nervous-, auditory-, endocrine-, or cardiovascular system, immunology and hematology, effects on the eyes, skin, behaviour, and dielectric heating of the whole animal. The reviews on potential carcinogenic effects and potential effects on reproduction and development are not discussed in this document, except when they also covered other exposure outcomes. Hence, the following review studies were not included in the metareview (Baan et al. 2011; Heynick and Merritt 2003; La Vignera et al. 2012; Vornoli et al. 2019). The environmental studies investigated reproductive, behavioural, or other effects. Out of those exposure outcomes that were investigated in this study, genotoxicity of RF-EMF exposure attracted most attention from review studies, with 17 studies focusing on in vitro genotoxicity and 16 on in vivo genotoxicity. Effects on the skin and endocrine system have been reviewed the least.

Some prior review studies only focused a limited number of potential outcomes of RF-EMF exposure. These are only discussed in the corresponding subsections. However, some review studies covered a larger spectrum of possible outcomes and also drew more generalized conclusions. These are discussed in general below and in detail in the appropriate subsections. In this section, all papers are summarized and discussed in alphabetical order.

(Cucurachi et al. 2013) investigated 113 studies on RF-EMF exposure of animals and plants. They analyzed the different studied effects of these papers and concluded that 'development and reproduction were the most studied ecological endpoints'. In their review, 56% of the studies on vertebrates found effects (irrespective of the endpoint). However, they did not find a dose-effect relationship for vertebrates.

(Habash et al. 2009) reviewed 45 studies on RF-EMF exposure of animals (in vitro and in vivo) that were executed between 2003-2007. They conclude that literature in general does not provide evidence of genotoxic effects of RF-EMFs at low levels, but they acknowledge that there are a few positive findings that need to be investigated further. They also state that some cellular studies provided evidence that gene expression is affected at RF-EMF exposure levels that were close to the safety limits in 2009 and that these studies should be followed-up. They stated that overall there is little evidence for cellular effects that are relevant for potential health effects below the RF safety limits.

(Heynick, Johnston, and Mason 2003) investigated at least 39 studies that investigated genotoxicity and carcinogenic effects of RF-EMF exposure of non-human vertebrates.

(Marino et al. 2011) reviewed both in vitro and in vivo studies on RF-EMF exposure with the aim of determining whether young vertebrates are more susceptible to potential effects induced by RF-EMF exposure. They reviewed 42 in vitro studies with 21 out of those studies that were focused on non-human vertebrates and 46 in vivo studies with different endpoints in vertebrate animals. They conclude that there is not enough information available in literature to determine whether there is an age-related sensitivity to RF-EMF exposure. They also conclude that 'dielectric heating remains the only established interaction mechanism that occurs at radiofrequencies'. However, they did not review any studies on dielectric heating.

(Obe 2004) reviewed papers published between 1990 and 2003 that investigated whether RF-EMF exposure could induce damage to the genetic material (assessed from DNA strand breaks, incidence of chromosomal aberrations, micronuclei and sister chromatid exchanges) of vertebrate cells. Out of those reviewed studies, 16 were in the category considered in this section. Limiting the review to non-human vertebrate animals, 7/16 of the reviewed studies showed increased genetic damage for RF-EMF exposed groups, while 7/16 did not show any increase in damage, and 2/16 were inconclusive.

(Panagopoulos and Margaritis 2008) reviewed a set of papers that investigated biological effects of non-thermal RF-EMF exposure of animals including vertebrates. However, they do not execute a systematic review and rather list some references without a clear inclusion criteria. (Panagopoulos, Johansson, and Carlo 2015; Panagopoulos 2019) reviewed some papers on RF-EMF exposure of animals including vertebrates in the context of a discussion of RF-EMF exposure setups and genotoxicity. However, not detailed outcomes of studies are presented.

(Repacholi 1997; 1998) executed two reviews: one focused on cancer and one focused on effects of low-level exposure. Both studies are a mixture of a meta-analysis and a classical review study. The review on RF-EMF and cancer included 48 papers, while the other review included approximately 100 papers (also including human studies).

(Vecchia 2009) is the largest publication that was reviewed in this section, covering nearly all topics that are studied in this field. They focused on publications after 1993 (because a previous review by the same organization was conducted in 1993) and reviewed 90 in vitro studies and 155 animal studies in the frequency range that is considered in this section. They provided the largest overview that can be found in any paper. However, they tend to be more critical for studies that find an effect of RF-EMF exposure at non-thermal levels, than for studies that did not find an effect.

Table 2 – Overview of review studies on RF-EMF exposure of vertebrates in the 0.4-6 GHz frequency band

Cellular Studies			Animal Studies										Environmental Studies			Nr studies	Reference
Genotoxicity	Non-genotoxic cellular effects	Cell transformation	Genotoxicity	Nervous system	Auditory system	Endocrine System	Cardiovascular System	Immunology & hematology	Skin	Eye	Behavior	Dielectric heating	Reproduction	Behavior	Other Effects		
																> 60	(Adair and Black 2003)
																< 20	(Balmori, Castilla, and Cortejoso 2006)
																< 20	(Balmori 2009)
																< 20	(Balmori 2014)
																< 20	(Balmori 2015)
																12	(Banik, Bandyopadhyay, and Ganguly 2003)
																>100	(Brusick et al. 1998)
																55	(Cotgreave 2005)
																113	(Cucurachi et al. 2013)
																11	(Deepinder, Makker, and Agarwal 2007)
																45	(Elder 2003)
																n/a	(Foster and Morrissey 2011)
																6	(Goodman, Greenebaum, and Marron 1995)
																6	(Gordon et al. 1963)
																45	(Habash et al. 2009)
																39	(Heynick, Johnston, and Mason 2003)
																>100	(Hossmann and Hermann 2003)
																>100	(IARC, 2013)
																n/a	(CNIRP, 2020)
																70	(Lai et al. 1987a)
																85	(Lin 2004)
																>160	(Manna and Ghosh 2016)
																16	(Obe 2004)
																25	(Panagopoulos and Margaritis 2008)
																70	(Marino et al. 2011)
																86	(Nittby et al. 2008)
																48	(Repacholi 1997)
																>100	(Repacholi 1998)
																22	(Repacholi et al. 2012)
																>100	(SCENIHR 2015)
																16	(Sienkiewicz, Jones, and Bottomley 2005)
																245	(Vecchia 2009)
																42	(L Verschaeve and Maes 1998)
																45	(L. Verschaeve et al. 2010)
																32	(Luc Verschaeve 2014)
																225	(Vijayalaxmi and Prihoda 2018)
																12	(Yu and Yao 2010)
																29	(Ziskin and Morrissey 2011)
17	12	8	16	14	6	2	4	4	1	3	6	7	4	5	3	Total Reviews on Outcome	

Cellular studies

Genotoxicity

(Brusick et al. 1998) reviewed more than 100 studies on the genotoxicity of RF-EMF exposure in the 0.8-3 GHz frequency range. They conclude that there is no direct evidence for mutagenic effects of RF-EMF exposure. They acknowledged that there might be some subtle indirect effects on replication and/or transcription of genes under some exposure conditions.

(Deepinder, Makker, and Agarwal 2007) list one study that showed damage to mitochondrial and nuclear genome in epididymal spermatozoa of mice under RF-EMF exposure.

(Habash et al. 2009) reviewed 13 studies on genotoxic effects of RF-EMF exposure, executed between 2003-2007. They concluded that at the time of their study (2009) there were still ongoing reports of possible genotoxic effects of RF fields. However, in their opinion the majority of the scientific evidence did not suggest that low-level exposure to RF fields induces genotoxic damage. They do advise to execute further research in this direction.

(Heynick, Johnston, and Mason 2003) reviewed eleven studies that investigated genotoxicity in non-human vertebrate cell (cultures) exposed to RF-EMFs in the considered frequency range. Almost all of them did not show a significant effect in comparison to sham exposure. The few studies that showed an effect either did not show a dose-relationship or were criticized for having confounding factors that were not related to RF-EMF exposure influencing the experiments.

(IARC 2013) reviewed a set of in-vitro studies with non-human cells that involved short-term, high-intensity exposures. These consistently gave positive results for DNA damage. In their opinion these were likely due to thermal effects. There were studies that showed effects and demonstrated the absence of effects in the subset of reviewed studies that were considered to be in the non-thermal range. They expressed a concern about some studies showing single-strand DNA breaks in vitro. However, they also pointed out that there were studies in their review that could not be replicated. There was one study in their review that showed altered microtubule structures at low exposures, which was concerning to the reviewers. In their conclusion, the authors considered the evidence for genotoxicity of RF-EMF as weak.

(Manna and Ghosh 2016) reviewed papers on in vitro RF-EMF genotoxicity in non-human and human vertebrates' cells. They found evidence in both directions in their review. They also investigated studies that look at genotoxicity of RF-EMFs in combination with another agent. These also showed contradictory results.

(Marino et al. 2011) reviewed 13 cellular studies on genotoxicity of RF-EMF exposure (human and non-human vertebrates) and found that in most of those studies there was no effect of exposure.

(Obe 2004) reviewed papers on RF-EMF genotoxicity (see section on animal studies and genotoxicity). The majority (10/16) of those studies were animal studies. In the cellular studies a majority (4/6) of the reviewed studies reported no increases in DNA damage for exposed cells.

(Panagopoulos and Margaritis 2008) report on two studies that investigated in vitro genotoxicity of RF-EMF exposure. Both studies did not show an effect.

(Repacholi 1997) reviewed studies on in vitro genotoxicity and concluded that most of those did not report any effects. (Repacholi 1998) concluded the same in his second review on low-level RF-EMFs. The majority of the in vitro studies that the author reviewed did not show genotoxic effects. In those studies that showed effects, temperature increases or secondary effects might be the underlying reason.

(SCENIHR 2015) stated that their previous review on in vitro genotoxicity had inconclusive results and that no dose-response had been demonstrated. They reviewed 31 additional studies on in vitro genotoxicity, 7 out of those investigated non-human vertebrates. 4/7 showed genotoxic effects.

(Vecchia 2009) reviewed 7 prior reviews (prior to 2003) on in vitro genotoxicity of RF-EMF. From those reviews they concluded that RF-EMFs are not directly mutagenic and that RF-EMFs probably do not enhance the genotoxicity of other agents in combined exposures. They reviewed eleven in vitro studies on genotoxicity of only RF-EMF exposure using the single cell electrophoresis assay. 5/11 found an effect of RF-EMF exposure (non-human vertebrate cells in the considered frequency range). They reviewed 9 in vitro studies on co-exposure to RF-EMF and other agents. 2/9 found an increased effect of RF-EMF in combination with another agent and 1/9 found an effect for RF-EMF exposure alone. The authors conclude that most studies included in their review, that also included human cells, have found no evidence of in vitro genotoxicity of RF-EMFs at non-thermal levels and that there is no additive effect for co-exposure to other agents. However, they do call for more research to clarify some of the positive effects that were seen. They acknowledge that not all studies were negative.

(L Verschaeve and Maes 1998) reviewed 9 in vitro studies on genotoxicity in non-human vertebrates. In four of those studies the genotoxicity of only RF-EMF exposure was studied and three out of those showed effects. Five investigated RF-EMF exposure in combination with another agent. None of those studies showed an added effect of the RF-EMFs in addition to the effect by the other agents. (L. Verschaeve et al. 2010) did a second review in which they reviewed 5 in vitro, cytogenetic studies of RF-EMF genotoxicity in non-human vertebrate cells. These studies did not find any genotoxic effects. One of the studies did find an effect on cell kinetics. The authors also focused on human cell lines and concluded that RF-EMF exposure does not induce cytogenetic damage, in particular not at non-thermal exposure levels. They also reviewed 4 in vitro studies of RF-EMF-induced DNA damage in non-human vertebrate cells. 2/4 studies found DNA damages. The authors contribute these findings to a thermal effect and potential issues with the data analysis, respectively. They reviewed 1 study on hamster cells that investigated γ -H2AX phosphorylated histone as a measure of RF-EMF-induced DNA damage. The study found an effect at some SAR levels, but did not find an effect on other levels. They reviewed 6 in vitro studies on combined exposures to RF-EMFs and chemical/physical mutagens. 4/6 found effects of the co-exposure and 1/6 found an effect of the RF-EMF exposure alone.

(Vijayalaxmi and Prihoda 2018) reviewed 225 studies on in vitro and in vivo genotoxic effects of RF-EMF exposure on mammalian cells. They conclude that the available data are inconsistent. Some studies found effects, while others have not. They also executed a meta-analysis where they weighed those effects that were shown in literature using quality control measures. The weighted outcome indicated a very small effect. They found a correlation between quality of the studies and reporting of no effect and an inverse correlation for those that reported increases in genetic damages. They also report on a publication bias towards those studies that found increases in genetic damages.

Cell transformation

(Habash et al. 2009) reviewed 10 studies on non-human vertebrates that investigated alterations in cellular functions. They found no evidence for an RF-EMF effect on cell cycle progression, proliferation and ornithine Decarboxylase (ODC) activity and no effect that low-level RF-EMFs might induce cell apoptosis.

(IARC Working Group on the Evaluation of Carcinogenic Risks to Humans, World Health Organization, and International Agency for Research on Cancer 2013) reviewed in-vitro studies focused on RF-induced apoptosis. All of them showed no effect except for one. They also state that the evidence that RF-EMFs alter cellular replication is considered weak (unclear whether this is for all vertebrates or only for humans).

(Manna and Ghosh 2016) reviewed a series of papers on effects of RF-EMF exposure on cellular morphology, proliferation, and growth profile. They do not draw any clear conclusions from that review. They also reviewed studies that investigated effects on cell death and cell cycle arrest induced by RF-EMF exposure, without any conclusions.

(Marino et al. 2011) reported on three studies that found that spontaneous neoplastic cell transformation of embryonic rodent cells was unaffected by RF-EMF exposure. They also reported on 8 studies that found no effect on apoptosis under RF-EMF exposure. Two studies reviewed by the authors on cell differentiation found contrasting results. One paper did not find any effect on mouse cell differentiation under RF-EMF exposure, while another paper found a derangement in chicken embryo retinal differentiation.

(SCENIHR 2015) reviewed six in vitro studies with non-human vertebrate cells on RF-EMF induced cell apoptosis executed between 2009 and 2015. Half of the studies demonstrated induction of apoptosis. They also reviewed seven in vitro studies that investigated effects of RF-EMFs on cell proliferation and cell cycle. 3/7 showed an effect, while 4/7 did not show an effect.

(Vecchia 2009) reviewed 14 studies that investigated a potential effect of RF-EMF exposure on cell proliferation, differentiation and cell cycle control. 9/14 studies reported an effect of RF-EMF exposure in at least one of the studied configurations and one reported on an effect of co-exposure with another agent. They also reviewed 3 studies that investigated a potential effect of RF-EMF exposure on apoptosis of non-human vertebrate cells. 1/3 found an effect on gene expression that is related to apoptosis. Nonetheless, the authors concluded that RF-EMF exposure has no effect on cell proliferation, cell cycle control, or on cellular apoptosis. 7 additional studies were reviewed on cellular transformation. 3 studies found an effect of a co-exposure to RF-EMFs and a promoting agent for cellular transformation. The authors concluded that there is no effect of RF-EMF exposure on cellular transformation.

(L Verschaeve and Maes 1998) reviewed 2 studies that investigated cellular transformation and cycle. Both studies found an effect of RF-EMF exposure.

Non-genotoxic cellular effects

(Banik, Bandyopadhyay, and Ganguly 2003) reviewed one study that found a change in membrane permeability in rabbit's red blood cells exposed to RF-EMFs.

(Cotgreave 2005) reviewed 55 studies that investigated whether RF-EMF exposure can influence the production of heat shock proteins (HSPs) or cause cellular stress in vertebrate cells (including human cells). Their review also included studies that investigated other potential changes in gene expression by RF-EMF exposure. They concluded that a number of in vitro studies have indicated that RF-EMF exposure can induce the expression of HSPs in a large variety of cell systems. It has also been demonstrated that modulation has an effect on this expression. However, the in vitro studies showed inconsistencies in exposure models, cell types used and the independent reproducibility of the findings. The authors questioned whether the effects can be described as non-thermal. The same effects have not been established with in vivo studies (contradictory results).

(Goodman, Greenebaum, and Marron 1995) report on changes in Ca^{2+} -efflux caused by modulated RF-EMF exposure of several cell types. They reviewed one study that reported on a decrease in the activity of the protein kinase C in lymphocytes under RF-EMF exposure at 450 MHz.

(Habash et al. 2009) reviewed 5 studies on non-human vertebrates where changes in genetic expression were investigated under RF-EMF exposure. The main hypotheses of the studies were whether RF-EMFs influence the expression of HSPs and immediate early genes (IEG). One of the reviewed studies found an effect on gene expression, one was inconclusive, and three did not find an effect. They did not find evidence in the reviewed literature that RF-EMFs might induce HSPs or a cellular stress response at low levels.

(Hossmann and Hermann 2003) reviewed studies on in vitro effects of RF-EMF exposure on neurons. Their review showed that at high SAR values (SAR 6.8–100 W/kg) isolated neurons respond to both continuous and pulsed RF-EMFs. They concluded that the modulation frequency has an influence on these in vitro effects. They attributed these effects to a thermal mechanism. One study was reviewed that investigated Ca^{2+} -efflux in nerve cells under RF-EMF exposure. Their review also investigated gene expression in rodents and showed that acute exposure of rats to low-level RF-EMFs did not activate HSPs.

(IARC 2013) reviewed almost 30 studies on expression of genes and protein changes in rodents exposed to RF-EMFs. They commented on the low quality of the studies and found mixed results in those studies that they considered of sufficient quality. The same review also investigated a set of studies on RF-EMF exposure in vivo with relation to the production of reactive oxygen species (ROS). They conclude that 'there was weak evidence that exposure to RF radiation affects oxidative stress and alters the levels of ROS'.

(Manna and Ghosh 2016) reviewed a series of studies that investigated HSP signaling and changes in gene expressions that are potentially induced by RF-EMFs. They did not draw any clear conclusions. They also investigated the production of ROS due to RF-EMF exposure on a cellular level and found studies with contradicting results.

(Marino et al. 2011) reviewed 15 studies (including human cells) that were focused on gene and protein expression under RF-EMF exposure. 9/15 studies did not find an effect of RF-EMF exposure. They reviewed one study on ROS formation in rodent cells, which found no effect. They reported on one paper that found an effect on activity of the enzyme Ornithine Decarboxylase (ODC) in exposed mouse cells. They reviewed 8 studies on the effects of combined exposures to RF-EMFs and other agents on a variety of outcomes. 3/8 studies found an effect.

(Repacholi 1998) reported on a series of effects on the cellular membrane. The author stated that several studies reported on RF-EMF fields influencing ionic channels formation, changes in frequency of channel openings, and increases in firing rates of those channels. No mechanism is provided and it is unclear whether these effects also lead to health effects. They also report on some changes in enzymes that are involved in signaling over the membrane under RF-EMF exposure.

(SCENIHR 2015) reviewed 4 studies focused on RF exposure and modification of the oxidation state of non-human vertebrate cells. These experiments commonly measured ROS-formation under RF-EMF exposure. In all (4/4) of the reviewed studies an increase in ROS formation was reported under RF-EMF exposure. One in vitro study in non-human vertebrate cells was reviewed that showed changes in protein expressions under RF-EMF exposure.

(Vecchia 2009) reviewed 6 studies that investigated the effect of RF-EMF exposure on Calcium ion (Ca^{2+}) metabolism (Ca-signaling) and ion channel dependent activity. Only 1/6 found an effect on the number of Ca^{2+} -spikes. 3 studies were reviewed on nitric oxide signaling in relation to RF-EMF (at 10 MHz) exposure. All three studies found effects of the exposure, but the reviewers criticize the dosimetry in those studies. One study found an effect on gap junction intercellular communication in rabbits and two studies found an effect of RF-EMF exposure on cell membrane receptor molecules. Two studies were reviewed that investigated the expression of particular genes (c-fos and c-jun). Both studies found an effect of RF-EMF exposure, but on one on c-fos and one on c-jun. One study was reviewed on non-human cellular transcriptomics. The study showed no effect of RF-EMF exposure. 3 studies were reviewed that investigated a potential effect of RF-EMF exposure on the production of ROS and oxidative stress. None of those found an effect. The authors concluded that the evidence of effects on calcium and nitric oxide signaling is very limited. They drew no conclusions on any effects on cellular gap junctions or cell membrane receptors. They also concluded that at the time of their review there was insufficient research to allow definitive

conclusions on gene expression and RF-EMF exposure. They conclude that RF-EMF exposure has no effect on ROS production.

(L Verschaeve and Maes 1998) reviewed 2 studies that investigated cancer-related cellular effects. Two studies showed an effect on intracellular levels of ODC, an enzyme usually implicated in tumor promotion.

Animal studies

Dielectric heating

(Adair and Black 2003) present an extensive review of the effects of RF-EMF heating and temperature increase in the body of several vertebrates. They state that for any given species, under any environmental conditions, an intensity of incident RF-EMF power density (the threshold value) can be determined that will reliably initiate a thermoregulatory response. They review a series of studies that determined these threshold values for non-human primates and smaller vertebrates (lab animals such as mice, rats, and rabbits) and studied the thermoregulatory responses of these animals (sweating, moving, respiratory changes, cardiovascular changes, etc.). They also presented a review of changes in metabolic heat production due to the added SAR. They reviewed the responses of several vertebrate animals to both prolonged RF-EMF exposure at thermal levels or short, intense exposure at thermal levels. A couple of studies were reviewed that investigated the effect of thermal heating using RF-EMFs in the early development of vertebrates. These showed that the thermoregulatory profile (and perhaps the metabolic response as well) depend(s) on age/developmental stage. They reported on some mixed results in growth rate of vertebrates exposed to thermal RF-EMFs during development. They reviewed three studies that investigated long-term (chronic) exposure to RF-EMFs at thermal levels, with some reported effects on body mass and oxygen intake. Finally, they reviewed some studies that investigated interaction between RF-EMF exposure and certain drugs.

(Foster and Morrissey 2011) reported on behavioural disruption in animals whose whole-body exposure corresponds to 4 W/kg, which in its turn is associated with a core temperature increase of 1°C. They also summarized a review on the relationship between whole-body SAR and body core temperature.

(Gordon et al. 1963) reported on dielectric heating and temperature increase of animals exposed to RF-EMFs at 10 mW/cm² and a reduction in endurance of animals exposed 10-40 mW/cm² (swim test).

(Lin 2004) listed a series of studies that investigated antenna configurations that could be used for hyperthermia treatment in animals and a second set of publications focused on RF-EMF ablation in dogs for cardiac surgery.

(Banik, Bandyopadhyay, and Ganguly 2003) list a series of studies that show dielectric heating of biological materials (including vertebrates) under RF-EMF exposure.

(Vecchia 2009) reviewed 3 studies on thermoregulatory responses in lab animals exposed to RF-EMFs. Those studies all found effects on metabolic heat production, heart rate, and blood pressure. The core temperature rose by 1°C in 2/3 studies. They stated that these changes are in line with the expected thermoregulatory changes to whole-body heating. These effects are independent of the heating method.

(Ziskin and Morrissey 2011) reviewed 19 studies that investigated thermal effects on development. Not all studies focused on hyperthermia induced by RF-EMF exposure. They investigated the relationship between maternal body core temperature and developmental abnormalities. They found that increases of more than 2°C above normal for extended periods of time, 2–2.5°C above normal for 0.5–1 h, or 4°C above normal for 15 min have resulted in developmental abnormalities in animal models. They referred to SAR values of more than 15 W/kg to reach such body core

temperature increases, with SARs above 4 W/kg corresponding to an increase of at least 1°C. There might be indirect effects through reduction in blood flow from mother to fetus at lower SARs, so they suggested a conservative limit of 1.5 W/kg. They also provided a limit on the localized SAR for the fetus. They also reported on, but did not present results of, three studies on RF-EMF exposure of vertebrate animals conducted between 2003-2010 that investigated developmental effects. They stated that these studies did not contradict previous reviews on the topic. They also reviewed 7 studies (2004-2010) that investigated effects of RF-EMFs on fertility in lab animals. 4/7 reported effects on fertility. However, the authors questioned the quality of the studies.

Genotoxicity

Some review studies on genotoxicity do not clarify whether those papers that are reviewed were animal or cellular studies. Those review papers are only discussed in one of the two sections on genotoxicity. Other reviews do explicitly divide their review in two parts (or only focus on one type), these are discussed in the appropriate sections.

(Brusick et al. 1998) reviewed more than 100 studies on the genotoxicity of RF-EMF exposure (see section on Cellular studies).

(Deepinder, Makker, and Agarwal 2007) discuss one study that showed damage to mitochondrial and nuclear genome in epididymal spermatozoa of mice under RF-EMF exposure. They also reviewed five studies on oxidative stress induced by RF-EMF exposure and conclude that it is debatable whether RF can induce oxidative stress.

(Habash et al. 2009) reviewed studies between 2003-2007 on genotoxic effects of RF-EMF exposure. This review included both cellular and animal studies (see section on cellular studies).

(Heynick, Johnston, and Mason 2003) reviewed eight studies on genotoxicity of in vivo RF-EMF exposure. These studies investigated single-and double-strand DNA breaks through assays of (parts) of the brain. These studies found significant differences in mean migratory length of the assays for some RF-EMF exposure conditions in comparison to sham, but no difference for other exposure conditions. Moreover, both the assay analysis and dosimetry were criticized in peer-reviewed literature.

(Hossmann and Hermann 2003) reviewed four studies on genotoxicity of RF-EMF with contradictory outcomes.

(IARC Working Group on the Evaluation of Carcinogenic Risks to Humans, World Health Organization, and International Agency for Research on Cancer 2013) reviewed a series of studies on genotoxicity of RF-EMF exposure of animals in vivo. However, they limited the review to studies in which they consider it proven that thermal confounding did not occur, where there was a clear reporting on the exposure conditions, and where sample sizes were large enough. Approximately half of the studies they found fell into that category. The remaining studies showed contradictory results.

(Marino et al. 2011) reviewed three in vivo studies on genotoxicity. One out of three showed an effect, while two did not show an effect.

(Obe 2004) reviewed papers on the genotoxicity of RF-EMF exposure on vertebrates. Out of those reviewed studies, 10 were on animal studies. 3/10 of the reviewed studies showed no increased damage for RF-EMF exposed groups, while 5/10 showed an increase in damage, and 2/10 were inconclusive.

(Panagopoulos and Margaritis 2008) discuss five studies that showed genotoxic effects due to (chronic) exposure to RF-EMFs and six studies that did not find any genotoxic effects.

(Repacholi 1997) reviewed two rodent studies that showed genotoxic effects. (Repacholi 1998) stated that several (3) rodent studies indicate that RF fields can influence DNA directly. (Repacholi et al. 2012) reviewed 10 in vivo studies on genotoxicity executed since 2000. 8/10 showed flaws in the study design and dosimetry. The two papers that satisfied all quality criteria did not find genotoxic effects.

(SCENIHR 2015) reviewed 5 in vivo studies on genotoxicity and concluded that there is evidence for such effects, but that better dosimetry is necessary in such studies.

(Vecchia 2009) reviewed 26 studies that investigated in vivo genotoxicity of RF-EMF exposure and co-exposure with another agent. 11 out of those studies found a genotoxic effect (1 of those in a co-exposure with another agent). Nonetheless, the authors concluded that most in vivo studies have failed to convincingly demonstrate any direct genetic effect after exposure of laboratory mammals to RF radiation.

(L Verschaeve and Maes 1998) reviewed 7 in vivo studies on genotoxicity of RF-EMF in vertebrates. Out of those 4 found effects and 3 did not find effects. (L. Verschaeve et al. 2010) reviewed 29 in vivo studies using laboratory mammals focused on genotoxicity of RF-EMF exposure or a combined exposure to RF-EMFs and another agent. They stated that 'many studies that have been published so far have not demonstrated convincingly direct DNA damage after acute or chronic exposure to RFR' while referring to 5 references. They then discussed 6 studies that did demonstrate that RF-EMFs can damage DNA in vivo. In an overview table, 13/29 studies found effects of RF-EMF exposure. The authors pointed out that there are replication problems with the studies that found effects.

Nervous and auditory system

(Foster and Morrissey 2011) summarized two reviews on the effect of RF-EMF heating on the nervous system. One of these reviews put forward a range of 0.5-5°C RF-EMF-induced temperature increase in the blood-brain barrier (BBB), which can cause a change in permeability. The second review discussed RF-EMF heating in the brain and what temperatures would lead to damages in the brain.

(Gordon et al. 1963) described a reduced sensitivity to acoustical stimuli in animals exposed to 3 GHz RF-EMFs. (Habash et al. 2009) reviewed six studies executed between 2004-2007 that investigated effects of RF-EMF exposure on the BBB in rodents. All studies used doses (≤ 6 W/kg) of RF-EMF and did not find an effect on the BBB.

(Hossmann and Hermann 2003) reviewed a series of studies that showed changes in EEG patterns in rodents and rabbits exposed to RF-EMFs. A second set of studies showed changes in the metabolism of the brain of rodents exposed to high-level RF-EMFs. They also list a series of studies that demonstrated auditory responses in animals, so-called microwave hearing. They also reviewed studies on the prevalence of molecules involved in neurotransmission in the brain under RF-EMF exposure. They only found pronounced effects for ELF exposure (not for RF). Finally, they presented a review of a series of studies on the BBB under RF-EMF exposure. Their review was inconclusive, but showed some evidence that BBB permeability increases (reversibly) at high SAR levels in some studies.

(IARC 2013) reviewed a series of studies that investigated permeability of the BBB under RF-EMF exposure. They found that one laboratory consistently showed an increase in the permeability of the BBB. However, the majority of studies in their review failed to observe any effect. Therefore, they classify the evidence that exposure to RF radiation alters the BBAs weak.

(ICNIRP, 2020) reported on studies on rodents and non-human primates that have shown decreases in food-reinforced memory performance during exposure to RF-EMFs at high whole-body averaged SAR (1°C increase in core temperature). They explain this as a thermal effect.

(Lai et al. 1987b) reviewed a set of 70 studies that investigated effects of RF-EMF exposure on the nervous system and compared those effects with those of psychoactive drugs. They also reviewed the effects of drugs on RF-induced hyperthermia and the influence of RF-EMF exposure on the effects of certain drugs. They highlighted the inherent difference between drug-administration, where drugs spread evenly over the body, and RF-EMF exposure which has morphology- and frequency-dependent absorption patterns. Their review showed some influence of RF-EMF exposure on the effect of barbiturates in rodents and rabbits. Some drugs were identified that counteracted hyperthermia and convulsions induced by RF-EMF exposure. Some other interactions with drugs were reported as well. They reported on the inexistence of an effect of low-intensity RF-EMF exposure on the BBB, while they state that higher intensities can change the BBB permeability and cerebral blood flow. They also report on sensory function that can be altered by RF-EMF exposure. They list a few studies that investigated the effect of RF-EMF exposure on neurotransmitter activity, but did not conclude on any effect. The authors also listed a series of studies that investigated the effect of RF-EMF exposure on the cholinergic system. Finally, they reviewed some studies that investigated the involvement of endogenous opioids in the vertebrate's response to RF-EMF exposure. They conclude that these opioids play a role in the effects of RF-EMF exposure.

(Lin 2004) reviewed a series of studies on changes in the BBB of rats under RF-EMF exposure. He stated that there exist several studies that show or do not show a change in the rat's blood-brain barrier's permeability at both high and low levels of RF-EMF exposure. The author attributed these mixed results to a lack of proper dosimetry in terms of SAR of the brain. Partial-brain exposure was suggested as a solution to this conundrum and used to demonstrate a dose-relationship of the effects of RF-EMF exposure on the change in permeability of the barrier. (Lin 2004) also reported on a series of studies that demonstrated microwave-hearing of pulsed high-peak RF-EMFs. These studies showed that RF-EMF pulses and acoustic pulses use the same pathway through the central auditory nervous system and that there lies a mechanical displacement at the base of this effect.

(Marino et al. 2011) investigated a set of papers that studied effects of RF-EMF exposure on brain structure, brain function, and the blood-brain barrier. They found mixed results in the studies investigating effects on brain structure and function. However, they found no evidence that exposure is associated with neural damage in developing brains. They also conclude that there is no strong evidence for permeability changes in the BBB. However, they single out a study in their conclusions that found cell losses in the cerebellum and hippocampus due to RF-EMF exposure. They stated that there are only a few studies that have investigated effects of RF-EMF exposure on hearing. They referred to two studies that found no effects on the cochlear function.

(Nittby et al. 2008) reviewed a series of papers that investigated effects of low-frequency and RF-EMFs on the BBB. They showed that studies on RF-EMF induced BBB disruption have shown contradictory results from different laboratories. Some groups demonstrated increased BBB permeability with their experimental conditions, whereas others did not.

(Panagopoulos and Margaritis 2008) listed 3 studies that investigated damage to brain cells due to changes in the blood-brain barrier that were induced by RF-EMF exposure. Two of those studies showed effects, while one did not show an effect.

(Repacholi 1998) stated that there are only a few studies that investigated effects of RF-EMF exposure on the nervous system at low-level exposure. Most studies used high-intensity RF-EMFs. The author reported on early studies at low exposure that did not show any changes in BBB permeability, but also listed two studies that did show effects at similar levels of exposure. Two reviews were cited that showed changes in electrical activity of the brain of cats and rabbits under exposure to RF-EMFs. Two studies were cited that show effects of RF-EMF exposure on neurotransmitters in the brain. Several studies that investigated microwave hearing were cited as well.

(SCENIHR 2015) reviewed a couple of studies (7) on RF-EMF exposure and BBB permeability. They concluded that RF-EMF exposure at SAR-values ≤ 2 W/kg causes impairment of the BBB. They also reviewed 3 studies that showed contradictory results in terms of RF effects on neurodegeneration. They reviewed six studies on ROS expression after in vivo exposure. Several of those studies suggested that RF-EMF exposure in rodents can cause oxidative stress effects. The study design of these studies was criticized by the reviewers. They also reviewed ten more in vivo studies that investigated other endpoints related to the neural system.

(Sienkiewicz, Jones, and Bottomley 2005) executed a review targeted at effects of ELF- and RF-EMF exposure on the nervous system. They stated that several studies have reported on effects on various neurotransmitter systems, but that at least some of those studies have been explained by temperature effects. Four studies were reviewed that showed effects of RF-EMF exposure on spatial memory. However, they also stated that four subsequent experiments failed to replicate those studies. Two studies did not find an effect on cognitive function of pre-natal RF-EMF exposure.

(Vecchia 2009) reviewed 7 studies related to gene expression in the nervous system of exposed lab animals. 3/7 studies found an effect. The authors started their review on effects of RF-EMF exposure on the BBB by summarizing past work. They stated that prior to 2000 several studies reported that low-level RF-EMFs may alter the permeability of the BBB, which would then cause negative effects. However, they stated that 'better conducted studies failed to corroborate these findings and the original observations were ascribed to various confounding factors'. Consistent changes in permeability were only found at relatively high SAR values (> 7 W/kg). They reviewed 11 studies on the topic, out of those four found an effect (3/4 conducted prior to 2000). The most recent studies did not find an effect. Therefore, the authors concluded that earlier reports of increased BBB permeability due to RF-EMF exposure have not been corroborated by later, better conducted studies. 9 studies were conducted on EEG under RF-EMF exposure. 8/9 studies showed an effect. The authors summed up a series of problems with the studies and concluded that it was not possible to draw any general conclusions regarding an effect of RF-EMFs on animal EEGs. 11 studies were reviewed on parameters related to neurotransmitters. All of these studies found an effect. Hence, the authors concluded that RF-EMF exposure might result in transient changes in these neurotransmitter related properties. However, they called for more studies to investigate whether these effects were caused by an auditory response or a heating effect. The authors also reviewed auditory effects of RF-EMF exposure. They started the review by stating that it is established that pulsed RF-EMFs can be perceived by lab animals (microwave hearing). However, it is not established that the modulation schemes used for telecommunication can induce such effects. The authors reviewed 4 studies that used GSM modulated and CW signals, these studies did not find any effect. They concluded that 'mobile phone type RF exposure has no effect on auditory function in rodents. It is also clear that animals can hear the pulsed RF characteristic of radar above given thresholds, through a thermoelastic expansion mechanism'.

Endocrine system

(Habash et al. 2009) reviewed two animal studies that investigated a potential effect of RF-EMF exposure on melatonin production. One did not find any effect, while the other one did find changes in melatonin production, which might be thermal in nature. (Vecchia 2009) reviewed 5 studies on potential effect of RF-EMF exposure on the endocrine system. One study reported an effect, while the other four studies did not find an effect.

Cardiovascular system

(Balmori, Castilla, and Cortejoso 2006) reported on changes in heart rhythms in toad hearts (*Xenopus Laevis*) that were exposed to RF-EMFs found in one study. (Gordon et al. 1963) describe a reduction in blood pressure of experimental animals exposed to 1 mW/cm^2 . (Panagopoulos and Margaritis 2008) discuss a study that showed an increased red blood cell count in exposed animals versus control. (Vecchia 2009) reviewed 7 studies on potential effects of RF-EMF exposure on the

cardiovascular system, including 3 that were aimed at studying a thermoregulatory response using dielectric heating (see section on dielectric heating). The remaining 4 studies did not find any effects on heart rate and $\frac{3}{4}$ did not find an effect on the blood pressure.

Immunology and hematology

(Banik, Bandyopadhyay, and Ganguly 2003) reported on changes in mammal's immunity induced by RF-EMF exposure. They reviewed seven studies that showed such effects. (Marino et al. 2011) stated that they found one good quality study on RF-EMF exposure and the immune system and that this study did not show any effects on the developing immune system. (Repacholi 1998) reported on a set of in vivo studies that showed effects on the immune system under RF-EMF exposure. However, those effects were similar to those that would occur under thermoregulation. (Vecchia 2009) reviewed 8 studies on immunology and hematology. Half of the reviewed studies found an effect. A previous review by the WHO in 1993 had concluded most effects on the immune system were transient and only occurred at high SAR levels. They concluded that the paper that they reviewed did not contradict that previous conclusion and most studies indicated that those changes in immune function and hematology that can be observed are transient and associated with temperature rise $\geq 1^\circ\text{C}$.

Skin

(Vecchia 2009) reviewed 5 studies on RF-EMF exposure of the skin. One found damages of the skin due to RF-EMF exposure and one found changes in the expression of certain genes. The methodology of these two studies was criticized by the authors.

Eye

(Elder 2003) reviewed 45 studies on ocular effects of RF-EMF exposure. They found that several ocular effects might occur under RF-EMF exposure. These are primarily cataracts, but can also be effects on the retina, cornea, and other parts of the eye. They reported on cataracts caused in rabbits' eye exposed at 2450 MHz for exposures of more than 30 min with very high localized SAR values ($\geq 150 \text{ W/kg}$). These SAR values were associated with temperatures ($\geq 41^\circ\text{C}$) in or near the lens of the rabbits. They also stated that studies with primates at similar levels did not result in the same incidence of cataract and hence question the potential to extrapolate the SAR results from rabbits to primates (including humans). They assumed that the same elevated temperature could induce cataract in the human eye. Very high whole-body exposures in rabbits did not induce cataract at non-lethal levels, while localized exposure can induce cataracts. They provided an overview of several studies that investigated phenomena in several parts of the eye, both using near-field RF-EMF exposure and far-field RF-EMF exposure. Long-term exposure studies using both monkeys and rabbits did not cause ocular effects.

(Foster and Morrissey 2011) reported on guidelines on localized exposure to RF-EMF, which are based on the occurrence of cataract in rabbits' eyes at a local SAR of 100 W/kg , which can cause a temperature increase up to 41.5°C at the level of the lens. They also presented a summary of another review that put forward a limit of 41°C for cataract on the lens. (Repacholi 1998) reported on a set of studies that showed effects of pulsed, low level RF-EMFs on the retina. The results could not be replicated in one CW study.

(Vecchia 2009) reviewed 5 studies on RF-EMF exposure and cataract development in the considered frequency range in this section. $\frac{3}{5}$ studies found an effect on the exposed animals' lens. They reported on an effect of anesthetics that reduce blood flow to the eye and therefore influence these studies. They reported on less sensitivity of primates to this effect than rabbits. They also reviewed 6 studies on RF-EMF exposure and effects on other tissues in the eye. $\frac{3}{6}$ found effects on the cornea using pulsed RF-EMFs, but these were not reproduced by authors from other labs. There were some reports on transient effects.

(Yu and Yao 2010) reviewed 4 studies that showed that high power RF-EMFs induce cataract in the lens. They reviewed 4 additional studies that investigated effects on lens transparency at non-thermal levels. One of those did not find any effects, while 3 found (reversible) effects. They report on 3 studies that found increased cell deaths in exposed lens epithelial cells (2 in vivo studies and one in vitro). They reviewed one study that showed an influence of RF-EMF exposure on gap junctional intercellular communication in RF-EMF exposed lens epithelial cells.

Behaviour

(Balmori 2009; Balmori, Castilla, and Cortejoso 2006) reported on adverse behaviour of rodents and rabbits exposed to RF-EMFs in a limited amount of studies. (Cucurachi et al. 2013) reviewed a series of lab studies on vertebrates (rats, mice, and rabbits) and analyzed changes in behaviour of those animals as a result of exposure. They concluded that the literature they reviewed presented contradictory results. (ICNIRP, 2020) reported on behavioural changes with the aim of reducing body temperature in non-human primates exposed to SAR levels that can induce temperature changes. (Marino et al. 2011) investigated six studies on animal behaviour under RF-EMF exposure and found two studies which showed improvements in performance (solution of maze). (SCENIHR 2015) reviewed 11 studies on learning, memory or behaviour under RF-EMF exposure. They found some studies that showed nonthermal effects, but also some studies that showed no effect. They comment on the low quality of the studies' RF-EMF exposure, blinding, proper controls, and dosimetry. (Vecchia 2009) reviewed 19 studies on animal behaviour under or after RF-EMF exposure. 13/19 studies found an effect. The authors attributed these effects to thermal effects or to auditory effects. They concluded that operant behaviour in laboratory rodents and primates can be disrupted by thermal RF exposure, which are sufficient to raise body core temperature by about 1°C. They were critical about those studies that showed an effect on non-thermal levels of exposure, but drew no clear conclusion.

Environmental studies

Behaviour

(Balmori 2009; 2014; 2015) reported on a negative correlation between the prevalence of house sparrows and electric field strength induced by the wireless network and changes in the activity of bats exposed to RF-EMFs. (Cucurachi et al. 2013) reviewed a limited number (< 5) environmental studies (dubbed field studies) that found a significant effect of RF-EMF exposure on breeding density and species composition in birds. There is overlap between the studies on birds reviewed in (Cucurachi et al. 2013) and (Balmori 2009; 2015; 2014).

(Luc Verschaeve 2014) reviewed studies that investigated the effect of environmental RF-EMFs on birds. They discussed a report that found it unlikely that communication towers cause disruption of night migrating birds' orientation or navigation systems. They also discussed one study that found no effect on homing success and vanishing time of pigeons (< 100 MHz RF-EMFs). However, they also discussed two studies that found an effect of low-level RF-EMFs on the geomagnetic orientation of birds. They also reviewed two studies on behavioural aspects of birds under RF-EMF exposure that are not related to orientation. Both studies found effects: one on aggression of birds and the other on the avoidance of exposure. They also reviewed a study in which the behaviour and survival of frogs was studied, while exposed to the RF-EMF telecommunication network. They criticized the experimental procedures used in the study. They reviewed one study that reported on severe behavioural changes in cows due to the placement of a broadcast tower. They reviewed two additional studies that found increased incidence of cataract in young cows that were exposed during development. Another study showed cytogenetic effect on the blood of cows that were exposed to a radar system. They reviewed two studies that investigated whether radar could be used to defer bats away from wind turbines. These studies showed that the prevalence of bats was lower on sites with lower RF-EMF intensity.

Reproduction

(Balmori 2009; 2014) reported a negative correlation between stork (*Ciconia Ciconia*) reproduction and exposure to RF-EMFs (electric field strength) emitted by the wireless network. (Cucurachi et al. 2013) reported on a limited number (< 2) of environmental study (dubbed field studies) that found a significant effect of RF-EMF exposure on reproduction of birds. (Luc Verschaeve 2014) discussed a study that found a reduced fertility in storks due to environmental RF-EMF exposure in Spain. They reviewed two studies that found a negative correlation between abundance of house sparrows and environmental RF-EMF field strengths. Another study was reviewed that did not show an effect on the nesting behaviour of tits near a radar installation. It is not clear whether this is a behavioural or reproductive effect. They reviewed one study that showed an effect on fertility of mice that were distributed around an antenna park. The study had a problem with the design of the control group.

Other

(Balmori 2015) reported on changes in the redox proteins and enzyme activities in cattle exposed to base stations at 900 MHz (Luc Verschaeve 2014) reviewed two studies that found a piezoelectric effect of RF-EMF exposure on bird feathers.

3.1.2. Review of effects on invertebrates

The literature review resulted in 122 publications on RF-EMF exposure of invertebrates in the targeted frequency range. Out of these, 15 were review papers, 7 were dosimetry studies, 25 only studied dielectric properties of invertebrates, and 3 studies were focused on insect monitoring using wireless sensor networks. This resulted in a set of 72 publications that are reviewed in this section. Out of those, 18 focused on dielectric heating using RF-EMFs, 44 were lab, experimental, or environmental studies that focused on insects, and 10 focused on other invertebrates. Figure 1 shows an overview of the study flow in this aspect of the review.

RF-EMF exposure of invertebrates in the 0.4-6 GHz frequency range was previously reviewed by (Cucurachi et al. 2013; Lin 2004; Newsom 1987; Panagopoulos and Margaritis 2008; Tanner and Romero-Sierra 1974; Válková and Vácha 2012; Luc Verschaeve 2014; Malkemper et al. 2018; Vanbergen et al. 2019). Additionally, there are several reviews on RF-EMF heating of invertebrates (Das, Kumar, and Shah 2013b; Diprose, Benson, and Willis 1984; Hou, Johnson, and Wang 2016; J. Johnson and Marcotte 1999; S. Wang and Tang 2001; Yadav et al. 2014).

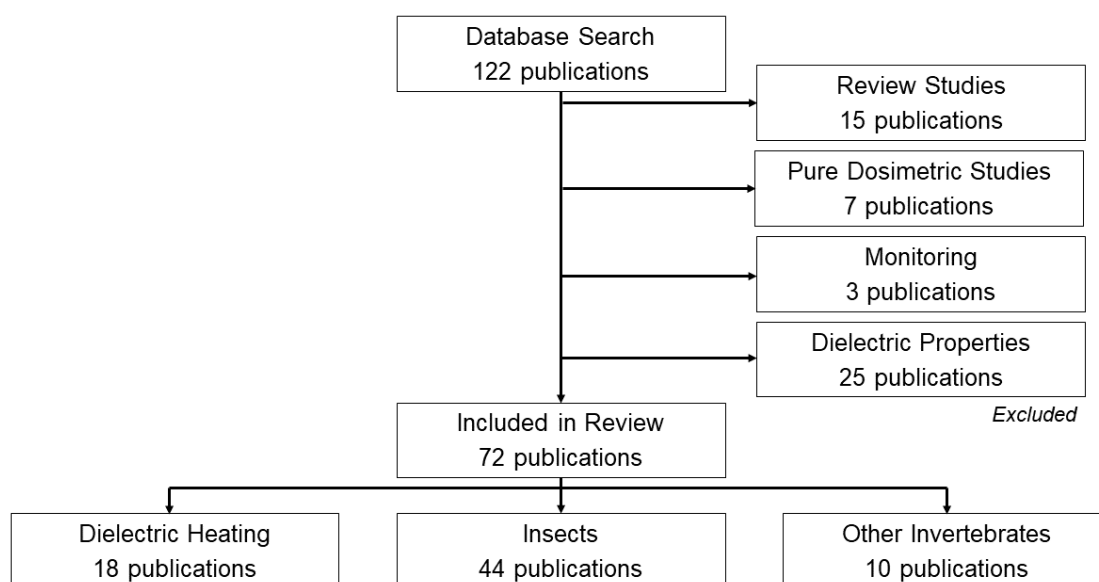


Figure 1 – Flowgraph of the post-processing of the literature review on low-frequency RF-EMF exposure of invertebrates

An important application of RF-EMFs in the studied frequency range is RF disinfestation of food materials or valuable wooden artefacts. This technique relies on differential dielectric heating between insects and the material they have infested. In order to estimate whether such an RF treatment is feasible, many studies have aimed at determining the dielectric properties of insects in the 0.4 – 6 GHz frequency band (J. Ahmed, Ramaswamy, and Raghavan 2007; Andreuccetti et al. 1995; Andreueeetti et al. 1994; Colpitts, Pelletier, and Cogswell 1992; Guo et al. 2011; Ikediala et al. 2000; Jiao et al. 2011; Rita Massa et al. 2014; Nelson and L. F. Charity 1972; Nelson 1966; 1996; Nelson and J. A. Payne 1982; Nelson et al. 1998; Nelson 1960; 2004; 1973; Nelson and Kantack 1966; Nelson and Stetson 1974; Nelson 1974; 2001; Nelson, Bartley, and Lawrence 1997; Ondráček and Brunnhofer 1984; Tanaka, Mallikarjunan, and Hung 1999; S. Wang, Tang, et al. 2003). These dielectric properties can then be used to determine the absorbed RF power or internal EMFs in the invertebrates. Additionally, they can be used for the design of RF-EMF exposure setups. Such studies are often called dosimetric studies. These are necessary input in studies that investigate the effects of RF-EMF exposure on invertebrates, but do not investigate such effects. Dosimetric results related to RF-EMF exposure of invertebrates can be found in (Ali and Al-Jabr 2003; Huang, Chen, and Wang 2015; Soproni et al. 2012; Thielens, Bell, et al. 2018; Thielens et al. 2020; Wang, J. Tang, et al. 2003; Fujiwara and Amemiya 1982). Finally, the review also resulted in a set of studies that use wireless networks, enabled by RF-EMFs, to monitor insect behaviour (Edwards-Murphy et al. 2016; Henry et al. 2019; Kridi, de Carvalho, and Gomes 2016). The studies referred to in this paragraph were not reviewed further in this section, but provide important input information for those studies that do investigate effects of RF-EMFs.

The reviewed studies are divided in three parts. First, studies that aim to investigate thermal effects of RF-EMF heating. Second, studies that aim to investigate non-thermal effects of RF-EMF exposure on insect species. Third, studies that aim to investigate neural responses in other invertebrate species.

Commonly investigated parameters are mortality of invertebrates in different life stages (this mainly applies to insects: egg, larva, pupa, adult), temperature changes, changes in water content, changes

in ELF-EMF potential on certain neurons under RF-EMF exposure, behavioural changes, genetic changes, and deformities or abnormalities during development.

Table 3 provides an overview of studies that investigated dielectric heating of invertebrates (insects) in the 450 MHz – 6 GHz frequency range. Most studies use 2.45 GHz as frequency of operation and a cavity to provide RF-EMF exposure to the insects. The majority of the references listed in Table 3 show mortalities up to 100% at the highest studied doses and show increases in mortality that scale with delivered RF-EMF dose. All of the studies listed in Table 3 demonstrate dielectric heating of invertebrates using RF-EMFs. The exposure levels of the studies listed in Table 3 are much higher than the exposure levels that can be expected in a real environment and also exceed the reference levels and basic restrictions put forward by the ICNIRP (ICNIRP, 2020).

In conclusion, Table 3 demonstrates that RF-EMFs in the 0.4-6 GHz range of very high intensity can lead to dielectric heating of insects and this heating can lead to high insect mortalities. Studies that investigate RF heating at frequencies below 400 MHz are not included in Table 3. However, it is very common that this heating is done at frequencies below 50 MHz (mainly 27 MHz) (Frings 1952; Hansen, Wang, and Tang 2004; Hansen, Drake, Heidt, et al. 2006; Hansen, Drake, Watkins, et al. 2006; Hansen et al. 2004; 2005; T. J. Headlee 1931; 1932; 1933; T. J. Headlee and Jobbins 1938; T. Headlee and Burdette 2020; Ikediala, J. Tang, and T. Wig 2000; Ikediala et al. 2002; Iritani and Woodbury 1954; Jiao et al. 2012; J. A. Johnson et al. 2004; J. A. Johnson, Wang, and Tang 2003; J. A. Johnson et al. 1998; A. M. Kadoum, Nelson, and Stetson 1967; Lowry et al. 1954; Mitcham et al. 2004; M.E. Monzon et al. 2006; Maria E Monzon et al. 2007; Rashkovan et al. 2003; Shrestha, Yu, and Baik 2013; S. Wang et al. 2001; S. Wang, Tang, et al. 2002; S. Wang, Ikediala, et al. 2002; S. Wang et al. 2007a; 2013; Webber, Wagner, and Pearson 1946).

Table 3 – Overview of papers that investigated RF heating of invertebrates (0.45-6 GHz)

Species	Frequency (GHz)	Exposure Conditions	Duration	Exposure Level or Input Power	Effect of RF-EMF Treatment	Reference
<i>Cryptolestes Ferrugineus</i>	2.45	Waveguide	≤ 40 s	600 W	Mortality up to 100 %.	(Hamid, Kashyap, and Cauwenberghe 1968)
<i>Cryptolestes Ferrugineus</i>	2.45	Cavity	≤ 56 s	≤ 500 W	Increased mortality after RF exposure. 100 % mortality at highest dose (56 s and 500 W).	(Vadivambal, Jayas, and White 2007)
<i>Cydia Pomonella</i>	0.915	Cavity	≤ 2 min	5 kW	Temperature increase up to 55°C. Mortality increased.	(Ikediala et al. 1999)
<i>Delia Radicum</i>	2.45	Cavity	≤ 40 s	≤ 6 kW	Increased mortality and temperature.	(Biron et al. 1996)
<i>Epehstia Cautella</i>	2.45	Cavity	≤ 150 s	900 W	90 s exposure is sufficient to result in 100 % mortality.	(Baysal et al. 1998)
<i>Hylotrupes Bajulus</i>	2.45	Open-ended waveguide	1 min	≤ 250 W	Heating up to 55°C and mortality up to 100%.	(Riminesi and Olmi 2016)
<i>Leptinotarsa Decemlineata,</i>	2.45	Waveguide	≤ 30s	≤1000 J/cm ²	Mortality increases with increasing dose. Reduced hatching of eggs with increasing dose.	(Colpitts, Pelletier, and Sleep 1993)
<i>Oligomerus Ptilinoides</i>	2.45	Open-ended waveguide	1 min	≤ 250 W	Heating up to 55°C and mortality up to 100%.	(Riminesi and Olmi 2016)
<i>Plodia Interpunctella</i>	2.45	Open-ended waveguide	≤40 min	≤ 150 W	Increased temperature and 100% mortality at 40 min exposure. Dose relationship is determined.	(Shayesteh and Barthakur 1996)
<i>Rhyzopertha Dominica</i>	2.45	Cavity	≤10 min	1 kW	Mortality up to 100 %.	(M. Ahmed et al. 2011)
<i>Rhyzopertha Dominica</i>	2.45	Cavity	≤ 26	1.6 kW	Heating up to 55°C. Increased mortality in comparison to unexposed groups. Combinations with gamma and infrared are studied as well.	(Kirkpatrick, Brower, and Tilton 1973)
<i>Rhyzopertha Dominica</i>	2.45	Cavity	≤ 25 s	2000 W	Mortality up to 100 % after 25 sexposure.	(Kirkpatrick and Roberts 1971)
<i>Rhynchophorus Ferrugineus</i>	2.45	Electrodes	≤ 35 min	1 kW	Heating up to 50°C.	(R. Massa et al. 2011)
<i>Sitophilus Granarius</i>	2.45	Waveguide	≤ 40 s	600 W	Mortality up to 100 %.	(Hamid, Kashyap, and Cauwenberghe 1968)

<i>Sitophilus Granarius</i>	0.9 & 2.45	Coaxial irradiation chamber	≤ 120 s	≤3 MW/cm ³	Mortalities up to 100 % at both frequencies.	(Ponomaryova, Riveray Oyarzabal, and Ruiz Sánchez 2008)
<i>Sitophilus Granarius</i>	2.45	Cavity	≤21 s	940 W	Increased mortality and temperature (> 100°).	(Baker, Wlant, and Taboada 1956)
<i>Sitophilus Granarius</i>	2.45	Cavity	≤ 56 s	≤500 W	Increased mortality after RF exposure. 100 % mortality at highest dose (56 s and 500 W).	(Vadivambal, Jayas, and White 2007)
<i>Sitophilus Oryzae</i>	2.45	Cavity	≤ 25 s	2000 W	Mortality up to 100 % after 25 sexposure.	(Kirkpatrick and Roberts 1971)
<i>Sitotroga Cerealella</i>	2.45	Cavity	≤ 25 s	2000 W	Mortality up to 100 % after 25 sexposure.	(Kirkpatrick and Roberts 1971)
<i>Sitotroga cerealella</i>	2.45	Cavity	25 s	unknown	Increased mortality which depends on age of insect. Combination with gamma radiation is investigated as well.	(Tuton et al. 1972)
<i>Tribolium Castaneum</i>	2.45	Cavity	≤ 56 s	≤ 500 W	Mortality increases with dose. Eggs were most susceptible, pupae the least.	(R Vadivambal, D S Jayas, and N.D.G White 2006)
<i>Tribolium Castaneum</i>	2.45	Cavity	≤ 56 s	≤ 500 W	Increased mortality after RF exposure. 100 % mortality at highest dose (56 s and 500 W).	(Vadivambal, Jayas, and White 2008)
<i>Tribolium Castaneum</i>	2.45	Cavity	≤ 56 s	≤ 500 W	Increased mortality after RF exposure. 100 % mortality at highest dose (56 s and 500 W).	(Vadivambal, Jayas, and White 2007)
<i>Tribolium Confusum</i>	2.45	Cavity	≤21 s	940 W	Increased mortality and temperature (> 100°).	(Baker, Wlant, and Taboada 1956)
<i>Tribolium Confusum</i>	2.45	Cavity	unknown	1.2 kW	Heating up to 65°C with mortalities up to 100 %.	(Hamid and Boulanger 1969)
<i>Tribolium Confusum</i>	2.45	Waveguide	≤ 40 s	600 W	Mortality up to 100 %.	(Hamid, Kashyap, and Cauwenbergh 1968)
<i>Tribolium Confusum</i>	2.45	Open-ended waveguide	≤40 min	≤ 150 W	Increased temperature and 100% mortality at 40 min exposure. Dose relationship is determined.	(Shayesteh and Barthakur 1996)

Those papers that study effects of RF-EMF exposure of insects are discussed for each insect type separately. In general, the lab studies investigating RF-EMF exposure (0.4-6 GHz) of insects that were found in literature, suffer from three general problems: (1) the quality of control and sham control groups or absence of control and/or sham, (2) quantification and stability of the RF-EMFs exposure, and (3) interference between effects due to RF-EMF exposure and other agents (sound, heating, and ELF exposure).

***Aedes Aegypti* (Yellow-Fever Mosquito)**

(Poh et al. 2017) investigated the behaviour (camera-tracked positioning) of *Aedes Aegypti* mosquitoes in an exposure chamber under RF-EMF exposure between 10 MHz and 20 GHz at an unknown exposure level. While the proposed measurement set up and study design in (Poh et al. 2017) is of great interest, the study does not provide any exposure assessment. Hence it is impossible to interpret what the actual exposure of the mosquitoes was. They did not find any difference in behaviour of the exposed groups in comparison to control and did not observe a reproducible frequency-dependency.

***Apis Mellifera* (Honey Bee)**

Several studies investigated RF-EMF exposure of *Apis Mellifera*, see Table 4. The first experiments in this frequency range were presented in (Westerdahl and Gary 1981; Gary and Westerdahl 1981), where bees were exposed to 2.45 GHz RF-EMFs with incident power densities of 3 - 50 mW/cm². They did not find changes in behaviour, sucrose intake, nor mortality between exposed groups and sham. However, the exposure of the sham control group was not determined in that study.

(Favre 2011) investigated the effect of the presence of a mobile phone on the sound produced by a bee hive (so-called piping). They were unable to determine RF-EMF exposure nor temperature in any experimental condition, which is problematic. They used a phone in stand-by mode as sham exposure. This sham exposure does not change sound of hive in comparison to unexposed control. The presence of an emitting phone, after 30 min of exposure, changed the sound of the hive. The effect could be thermal (no temperature measurements) and was reversible.

(Vilić et al. 2017) exposed honey bees in a TEM cell at 900 MHz at different levels of exposure. They did use a sham exposed group, but did not measure the exposure of that group. They investigated oxidative (stress) response and genotoxicity and found some significant differences between sham and exposed groups at some exposure levels for some studied parameters. They did not find a consistent effect over all exposure levels or a dose-response.

Honey bee exposure to RF-EMFs was also studied in (Halabi, Achkar, and Haidar 2013; Kimmel 2007; Lopatina et al. 2019; V. P. Sharma and Kumar 2010), but these studies suffer from significant experimental flaws, such as: absence of sham (V. P. Sharma and Kumar 2010; Kimmel 2007; Halabi, Achkar, and Haidar 2013) and no determination of exposure level with sham that differs from unexposed control (Lopatina et al. 2019).

Table 4 – Studies that investigate RF-EMF exposure (0.4-6 GHz) of *Apis Mellifera* (honey bee)

Frequency (GHz)	Exposure Conditions	Duration	Control	Sham	Exposure Level	Effect of RF-EMF Exposure	Reference
0.9	Mobile phones	< 20 h	Control with inactive phones and phones in 'standby' mode. Exposure of control was not measured. Sound was measured. Unexposed control is also used.	yes	Not determined	Sham does not change sound of hive (piping) in comparison to unexposed control. After 30 min of exposure the sound of the hive changes. Effect could be thermal (no temperature measurements).	(Favre 2011)
0.9-2.2	Mobile Phone	15 min/day	no	no	Not determined	Effect on sound of bee hive after 12 min of exposure. Decrease of hive size after prolonged exposure.	(Halabi, Achkar, and Haidar 2013)
2.45	Horn antenna in exposure chamber	30 min	3 controls: sham, lab control, and hive control. Exposure of controls was not measured.	yes	3 - 50 mW/cm ²	No behavioural effects were found.	(Gary and Westerdahl 1981)
1.9	DECT Station	unclear	Shielded control. Exposure of control was not determined.	no	2.5 mW transmitted power	No difference between exposed and unexposed groups in an index that studies return to hive.	(Kimmel 2007)
2.45	Wi-Fi Access Point in faraday cage	2-24 h	Unexposed control and two sham control groups. Exposure was not measured for any group.	yes	unclear	Reduced incidence of unconditioned and conditioned feeding response in exposed insects (conditioned response also altered by sham).	(Lopatina et al. 2019)
0.9	Mobile phone	15 min, twice/day, for up to 1500 h	Unexposed control and 'sham' with dummy phones. Exposure of control and sham was not measured.	no	56.8 V/m (measured)	Changes in foraging behaviour after exposure. Changes in colony size after exposure. Small sample size and no statistics were used.	(V. P. Sharma and Kumar 2010)
0.9	Mobile phone	30 min/day, two weeks total	Control group is sham exposed. Exposure of sham was not measured.	yes	0.9-3.8 V/m (measured on one instance)	Reduced chances of queen survival in exposed groups. Decrease in hatching of queens. No change in mating success. No changes in colonies.	(Odemer and Odemer 2019)
0.9	TEM cell	2 h	Control group was sham exposed. Exposure of sham control was not measured.	yes	10, 23, 41 and 120 V/m	Oxidative response and genotoxicity were investigated. Some significant differences between sham and exposed were observed at some levels or some studied parameters, but were not seen at other exposure levels.	(Vilić et al. 2017)
2.45	Exposure chamber	0.5-24 h	Unexposed control and group in sham chamber. Exposure of control and sham were not measured.	yes	3-50 mW/cm ²	No changes in consumption of sucrose and mortality.	(Westerdahl and Gary 1981)

Table 5 – Studies that investigate RF-EMF exposure (0.4-6 GHz) of *Drosophila Melanogaster* (fruit fly)

Frequency (GHz)	Exposure Conditions	Duration	control	sham	Exposure Level	Effect of RF-EMF Exposure	Reference
1.9	Mobile phone	60 min, twice per day, 10 days	Control was exposed to phone with power off. Exposure of control was not measured.	yes	1.5 - 3.3 V/m (measured). ELF-EMFs are measured as well.	increased numbers of offspring (adults and pupae). Cellular effects: elevated hsp70 levels, increased serum response element, DNA-binding and induced the phosphorylation of nuclear transcription factor ELK-1.	(Weisbrot et al. 2003)
0.8	Patented exposure device (cavity)	< 36 h	Control were unexposed insects. Exposure of control is not measured.	no	1.6 – 4 W/kg	High exposure group has reduced viability after 18 h of exposure. Low exposure group after 36 h of exposure. RF-EMF exposure triggers cellular stress response and certain signaling responses.	(Lee et al. 2008)
2.45	Antenna	6 h	Unexposed control and control immersed in water. Negative control with X-rays. Exposure not measured.	no	100 W/kg.	No mutagenic activity due to RF-EMF exposure. Difference with X-ray exposure.	(Hamnerius et al. 1979)
0.03-3	Electrodes, Helmholtz coil, cavity, horn antenna	6 h	Control were unexposed insects. Exposure of control is not measured.	no	0.3 W/kg (27 MHz), 110 W/kg (2.45 GHz), 60 W/kg (3 GHz)	None of the RF-EMF exposures gave an elevated mutation frequency.	(Hamnerius, Rasmuson, and Rasmuson 1985)
0.8-1.9	Mobile phone and DECT phone	20min/day	Control were unexposed insects. Exposure of control is not measured.	no	Calculations are presented	No convincing effect on reproductive capacity. Paper was criticized for not having sham exposure.	(Geronikolou et al. 2014; Dimitris J and Andreas 2020; Geronikolou et al. 2019)
0.9-1.8	Mobile phone	<6 min/day	Control group was exposed to phone in off-mode. Exposure of control was not measured.	yes	0.35 ± 0.07mW/cm ² (measured)	Reduced number of pupae per maternal fly for exposed groups. Elevated incidence of egg chambers with fragmented DNA or disorganized actin network.	(Chavdoula, Panagopoulos, and Margaritis 2010)
1.9	Duct access point	0.5, 1, 6, 24 and 96 h	Unexposed Control (shielded) and sham control. Exposure of control and sham were not measured.	yes	2.7 V/m (measured)	Reactive oxygen species (ROS) cellular contents were found to increase for exposures longer than 6 h. This response was present for shorter exposures in the ovaries of female flies. No difference between sham and control.	(Manta et al. 2014)

1.8	Mobile phone	30 min	Sham control: switched off mobile phone. Exposure of sham control is not measured.	yes	10 V/m (measured)	Reactive oxygen species (ROS) cellular contents were found to increase. Some changes in gene expression.	(Manta et al. 2017)
2.4	waveguide	5-60 min/day; longer exposure is at lower level for 1 to 5 days.	Control is untreated sample. Exposure of control is not measured	no	15-25 W/cm ²	Increasing days of exposure decreased the survival rate. Highest power level also causes an additional mortality in comparison to lower levels. No effect on sex ratio of offspring, but reduced numbers for the longer exposure.	(Marec, Ondráček, and Brunnhofer 1985)
2.45	waveguide	10 min	Unexposed control and sham control. Exposure of control and sham are not measured. Control with alternative heating method.	yes	0.644 W/g (calculated)	Reduced number of eggs per female of RF exposed group in comparison to sham and control. RF exposure did result in heating. Alternative heating method produces a similar reduction in eggs per female. Lower survival of eggs for RF-EMF exposed groups in comparison to sham, control, and alternative heating method.	(Pay, Andersen, and Jessup 1978)
0.029 and 0.15	Near Field of antenna	12 h	Control is untreated sample. Exposure of control is not measured	no	62 V/m (150 MHz) and 600 V/m (29 MHz), measured	No increase in tested genetic aberrations in offspring of exposed or unexposed flies.	(Mittler 1976)
0.02-2.4	A set of RF Devices	Various exposure schemes. Up to 7 d of exposure, up to one hour per day.	Unexposed control and so-called sham. Sham was not exposed to non-emitting device. Sham was shielded. Exposure of sham was monitored. ELF was measured as well.	no	0.3 -22 V/m (technology dependent, measured)	Increased percentage of ovarian apoptotic follicles. Reduced fecundity (viable eggs/female). Both quantities are correlated. A dose-relationship is demonstrated using different exposure times and separation distances.	(Margaritis et al. 2014a)
0.1-0.9	Antenna	6 or 60 min/day for 6 days, or 6 or 60 min on the 6 th day	Unexposed, shielded control.	no	0.2-9 V/m	Increases in apoptotic cell death in comparison to control for most of the exposure groups. There are significant but smaller differences between control groups.	(Sagioglou et al. 2016)
0.9	Mobile phone	6 min every 10 h	Sham control and unexposed control. Exposure of control and sham were not measured.	yes	0.354 ± 0.063 mW/cm ² (measured). ELF-EMFs also measured.	Change in ovarian size of exposed groups after 20 h of exposure. This is attributed to DNA damage by the authors	(Panagopoulos 2012)

0.9	Mobile phone	6 min/day, <5 days	Two sham exposed controls. Exposure of sham was not measured.	yes	0.436+/-0.060 mW/cm ² (data transmission); 0.041+/-0.006 mW/cm ² (low data transmission). ELF is also measured.	Decreases in the reproductive capacity (number of pupae per maternal fly). Effect of usage of the mobile phone (high or low amount of data transmitted) is observed.	(Panagopoulos, Karabarounis, and Margaritis 2004)
0.9-1.8	Unclear	6 min/day, <5 days	Sham control. Exposure of sham was not measured.	yes	0.4 mW/cm ² (measured)	Increased ovarian DNA fragmentation in comparison to sham and ELF exposure.	(Panagopoulos 2019)
0.9-1.8	Mobile phone	6 min/day, 5 days	Sham control. Exposure of sham was not measured.	yes	0.4 mW/cm ² (900 MHz, measured), 0.3 mW/cm ² (1800 MHz, measured). ELF also measured.	Decreases in the reproductive capacity after RF-EMF exposure.	(Panagopoulos, Chavdoula, Karabarounis, et al. 2007)
0.9-1.8	Mobile phone	6 min/day, 5 days	Sham control and unexposed control. Exposure of control and sham were not measured.	yes	0.4 mW/cm ² (900 MHz, measured), 0.3 mW/cm ² (1800 MHz, measured). ELF also measured.	Increases in ovarian cell death after RF-EMF exposure.	(Panagopoulos, Chavdoula, Nezis, et al. 2007)
0.9-1.8	Mobile phone	6 min/day, 5 days	Sham control and unexposed control. Exposure of control and sham were not measured.	yes	0.004-0.4 mW/cm ² (ELF exposure also measured)	Decreases in the reproductive capacity and increases in ovarian cell death after RF-EMF exposure up to a certain separation distance from the mobile phone (dose-relationship).	(Panagopoulos and Margaritis 2010)
1.8-2.7	Antenna	12h/day, 5 days	Sham control in anechoic chamber. Exposure was not measured, but sham was shielded during normal exposure.	yes	29 mW/m ² (calculated)	Eight properties of the brain were studied. Only one of those properties showed a significant change for exposed in comparison to sham (Euler number).	(A. Singh et al. 2020)

Table 6 – Studies that investigate RF-EMF exposure (0.4-6 GHz) of ants

Species	Frequency (GHz)	Exposure Conditions	Duration	control	sham	Exposure Level	Effect of RF-EMF Exposure	Reference
<i>Myrmica sabuleti</i> (ant)	0.9	Patch antenna	Several days (intermittent)	Control were unexposed ants. Exposure of control is not measured.	no	0.77 V/m (calculated)	Reduced efficiency in olfactory and visual conditioning. Increased memory loss.	(M.-C. Cammaerts et al. 2012)
<i>Myrmica sabuleti</i> (ant) and <i>Myrmica ruginodis</i> (ant)	0.9-2.4	Mobile phone, Smartphone, DECT phone, Wi-Fi Access point, and Laptop	Unclear. For the duration of the experiment.	Sham exposure with devices off and blind unexposed control. No measurements of exposure are presented.	yes	unclear	Some effects on linear and angular speed of the ants. However, the sham groups also showed a difference in comparison to the control.	(M.-C. Cammaerts and Johansson 2014)
<i>Myrmica sabuleti</i> (ant)	0.9	Patch antenna	2.5 days	Control is unexposed group. Exposure of control is not measured.	no	0.77 V/m (calculated)	Ants' response to certain pheromones was studied. Also, a potential effect on food collection was investigated.	(M.-C. Cammaerts et al. 2013)
<i>Myrmica sabuleti</i> (ant)	0.94	Log-periodic Antenna	10 min	Background exposure is measured (0.024 V/m). Control group is exposed to these levels.	no	1.5 V/m (and two levels at 10 dB and 50 dB lower)	Behavioural effects were observed. Ants' reaction to certain pheromones was altered.	(M.-C. Cammaerts, Vandenbosch, and Volski 2014)

Dermacentor Reticulatus (tick)

The movement of *Dermacentor Reticulatus* exposed to RF-EMFs at 900 MHz was studied by (Vargová et al. 2017). A power density of $700 \mu\text{W}/\text{m}^2$ (calculated, not measured) was used during 4 minutes. An increased movement of the insects was observed during exposure in comparison to the same insects when they were not exposed. The background exposure during the periods without RF-EMF exposure should be low because the tests were executed in an anechoic chamber

Drosophila Melanogaster

Table 5 lists those studies that have investigated effects of RF-EMF exposure on *Drosophila Melanogaster*. There were a series of studies in the 1970-80's that investigated exposure of drosophilae to RF-EMFs. Several of those, executed at frequencies 0.1-2.45 GHz found no additional genetic damages in the exposed flies in comparison to unexposed control (Hamnerius et al. 1979; Hamnerius, Rasmuson, and Rasmuson 1985; Mittler 1976). However, these studies did not have a sham exposed group and did not measure the exposure of their unexposed control. (Marec, Ondráček, and Brunnhofer 1985) did not observe any genetic effects after exposure of drosophilae to 2.4 GHz RF-EMFs. However, they also did not use a sham exposed group. The paper does present some effects on reduced survival rates at higher exposure levels ($25 \text{ W}/\text{cm}^2$) in comparison to lower exposure levels ($\leq 20 \text{ W}/\text{cm}^2$).

In the same time period, (Pay, Andersen, and Jessup 1978) investigated RF heating of drosophila at 2.45 GHz in comparison to a sham and an unexposed control group. They observed a reduced number of eggs per female in the RF exposed group in comparison to sham and control. An alternative heating method produced a similar reduction in eggs per female. However, they also observed a lower survival of eggs for RF-EMF exposed groups in comparison to sham, control, and alternative heating method. This effect could not be explained by the elevated temperature alone.

In more recent work, a Greek research center has published a large set of studies that demonstrated effects of RF-EMF exposure (0.8-2.5 GHz) of *Drosophila* (Chavdoula, Panagopoulos, and Margaritis 2010; Manta et al. 2017; 2014; Margaritis et al. 2014b; Sagioglou et al. 2016; Panagopoulos 2012; Panagopoulos, Karabarbounis, and Margaritis 2004; Panagopoulos 2019; Panagopoulos, Chavdoula, Nezis, et al. 2007; Panagopoulos, Chavdoula, Karabarbounis, et al. 2007; Panagopoulos, Chavdoula, and Margaritis 2010). These studies are faced with some major experimental issues as well. Most of their studies use actual RF-EMF emitting devices as sources. This implies that there is no control on the RF-EMF exposure values, since the network operator determines the output power of a device. The studies present power density and electromagnetic field values, which have been criticized (Luc Verschaeve 2014), because the RF-EMF exposure levels are measured incorrectly. The exposure values are also measured in one-time instance, instead of continuously throughout the experiment. One of their publications does use a signal generator and antenna with controlled output power as RF source (Sagioglou et al. 2016). However, this particular study lacks the presence of a sham exposed group. In another publication, the problem of inaccurate exposure assessment is circumvented by using different separation distances (Margaritis et al. 2014b). However, this paper also does not have a real sham exposed group. Apart from (Sagioglou et al. 2016; Margaritis et al. 2014b) the studies from this research group have the main advantage that they have a sham exposed group as control. This aspect was lacking in the state-of-the-art prior to their research. However, exposure of that sham group (or the unexposed control) was never measured. They reported a series of effects in drosophilae after RF-EMF exposure in comparison to sham: reduced number of pupae per maternal fly (Chavdoula, Panagopoulos, and Margaritis 2010; Margaritis et al. 2014a; Panagopoulos, Karabarbounis, and Margaritis 2004; Panagopoulos, Chavdoula, Nezis, et al. 2007; Panagopoulos, Chavdoula, and Margaritis 2010), increased reactive oxygen species in cellular contents (Manta et al. 2017; 2014), (DNA) problems with ovarian cells (Margaritis et al. 2014b; Panagopoulos 2019; Panagopoulos, Chavdoula, Karabarbounis, et al. 2007; Panagopoulos, Chavdoula, and Margaritis 2010), Increases in apoptotic cell death (Sagioglou et al. 2016), and

changes in ovarian size (Panagopoulos 2012). All of those are related to reproductive problems caused by RF-EMF exposure.

(A. Singh et al. 2020) study the effect of RF-EMF exposure at 1.8-2.7 GHz at 29 mW/m² on parameters of the brain of drosophilae. They investigated eight parameters and found changes in one of those parameters in comparison to sham exposure (shielded sham).

There are also a couple of recent studies that investigated RF-EMF exposure of drosophilae without sham group and/or exposure assessment of the control group (Geronikolou et al. 2014; 2019; Dimitris J and Andreas 2020; Lee et al. 2008), see Table 5. (Geronikolou et al. 2014) did not find an effect on the reproductive capacity. (Lee et al. 2008) found a dose-related effect on insect viability and found that RF EMF exposure triggers cellular stress and certain signaling responses.

When considering drosophila, the literature is seriously flawed. Studies either do not use sham control or when they use sham control, they provide unreliable exposure measurements. It seems that those studies that have a sham exposed group did find significant effects of RF-EMF exposure, while those relying on an unexposed (potentially exposed) control group did not find any effects. Almost no studies provided measurements of the exposure of the sham or control groups. This makes an interpretation of their results very difficult.

Myrmica Sabuleti

A series of papers from one research group has investigated the effects of RF-EMF exposure on ants (*Myrmica Sabuleti* and *Myrmica Ruginodis*) (M.-C. Cammaerts et al. 2012; M.-C. Cammaerts and Johansson 2014; M.-C. Cammaerts et al. 2013; M.-C. Cammaerts, Vandenbosch, and Volski 2014). Only one of their studies involves a sham control group (M.-C. Cammaerts and Johansson 2014). In that study the sham showed a significant difference in comparison to the unexposed control in the studied effect. The RF-EMF exposure is only measured in one of their studies (M.-C. Cammaerts, Vandenbosch, and Volski 2014). Moreover, some of the exposure conditions involve exposure to other agents such as hot air displacement, sound, and ELF-EMFs (Luc Verschaeve 2014). Their studies are focused on behavioural aspects of the ant colonies, conditioning, and retention of conditioned responses. They demonstrate behavioural changes and changes in conditioning in comparison to their control groups, but the exposure of the control is unknown.

Table 7 – Studies that investigate RF-EMF exposure (0.4-6 GHz) of *Tenebrio Molitor* (beetle)

Frequency (GHz)	Exposure Conditions	Duration	control	sham	Exposure Level	Effect of RF-EMF Exposure	Reference
4-6	Horn Antenna and reflector	5 min- 6 h	Sham control. (in shielded chamber). Control with other heating method.	yes	38-1526 J/kg (< 24 mW/cm ²)	Increases in mortality and deformities under RF EMF exposure. Temperature increase up to 15° was measured. Effect of other heating method produces lower mortalities and deformities at similar temperature difference.	(Olsen 1977)
6	Horn Antenna and reflector	1.5-24 h	Unclear	unclear	1123 J/g	High SAR (208 W/kg) and short exposure produces deformities and mortality, while lower SAR (13 W/kg) and longer exposure time (same dose) produces no deformities or additional mortality. They find a polarization difference. Temperature increase is measured.	(Olsen 1982)
6	Horn Antenna with(out) reflector	2 -13 h	Unclear how the control is performed	unclear	130 W/Kg and 54 W/kg (polarization)	Effect on number of mortalities and deformities in the exposure case where the magnetic field was parallel to the insect with reflector. No effects in free space and E-field parallel to pupae.	(Pickard and Olsen 1979)

Tenebrio Molitor

A series of papers investigated RF-EMF exposure of the beetle *Tenebrio Molitor*, see Table 7. (Pickard and Olsen 1979; Olsen 1977; 1982) studied exposure conditions with relatively high SAR values and also measured temperature increases at these SAR levels (Olsen 1977). They find increased deformities and mortality at exposure to intense RF-EMFs. These effects depend on the delivered SAR, rather than on the delivered dose (Olsen 1982). They also find an effect of polarization (Pickard and Olsen 1979) and compare to an alternative heating method that induces the same temperature increase, but not the same effects on mortality and development (Olsen 1977).

Environmental studies

The studies on invertebrates that are discussed above use experimental setups to generate RF EMF exposure. An alternative study protocol uses the RF-EMF exposure that is present in the environment to investigate potential effects of that exposure on invertebrates. In the case of insects, this approach was first described in (Mittler 1977). They investigated two groups of *Drosophila melanogaster* either exposed or not exposed to a radio broadcast tower (100 MHz). The exposed group experienced an incident field strength of 0.3 V/m (Mittler 1977). No effects were found in the tested genetic aberrations. Unfortunately, the exposure of the control group was not measured, so the group might have been exposed to the same RF-EMFs (radio broadcasts commonly cover wide areas). However, the study's attempt to investigate realistic exposure scenarios is valuable. (Pramod and Yogesh 2014) used a similar protocol to investigate the effect of 900 MHz RF-EMFs emitted by a base station on *Apis Mellifera* (honey bee) colonies. They used three study groups: one at the base of the tower (0.35 V/m), one equipped with mobile phones (57 V/m during calls) at 2 km from the tower (a proxy for a user), and one at another site with low RF-EMF exposure (7 mV/m) without any device as control. The study could have benefitted from a fourth group with sham exposure and of course the exposure generated by the phones could not be controlled by the investigators. However, again the attempt at obtaining realistic field exposure has its merits. They did not find an effect on the hives' brood area when comparing the 3 groups. (Vijver et al. 2014) investigated a set of insects: Springtails (*Folsomia Candida*), predatory bugs (*Orius Laevigatus*), parasitic wasps (*Asobara Japonica*), and fruit flies (*Drosophila Melanogaster*). These were placed for 48 h in an outdoor environment that was covered by a 900 MHz base station antenna (the telecommunication network). RF-EMF exposure was measured on each site where insects were placed and a shielded control group was placed on the same location (at 2 m from the exposed insects). Reproductive parameters were studied and no effects were found. An alternative study approach was used in (Lázaro et al. 2016). Instead of investigating a specific species and bringing samples of that species into an exposure conditions, the authors of (Lázaro et al. 2016) used an insect trap to collect several types of wild pollinators at different distances from telecommunication base station antennas (0.8-2.6 GHz) on two Greek islands. The electric field strength was measured on each experimental site (not during the entire experiment) and correlated with the abundance of different groups of pollinators. Contrasting effects were obtained on different groups of pollinators.

Table 8 – Studies that investigate RF-EMF exposure (0.4-6 GHz) of non-insect invertebrates

Species	Frequency (GHz)	Exposure Conditions	Duration	control	sham	Exposure Level	Effect of RF-EMF Exposure	Reference
<i>Aplysia (sea slug)</i>	1.5-2.45	Microstrip line. Dissected ganglia.	Minutes	Unexposed sample.	unclear	0-50 mW/cm ³	Effect on neural firing is observed even below 10mW/cm ³ .	(Wachtel, Seaman, and Joines 1975)
<i>Aplysia Californica (sea slug)</i>	1.5-2.45	Microstrip line. Dissected ganglia.	< 3 min	Unexposed sample.	unclear	0-80 mW/g	Dose-related effect on neural firing.	(Seaman and Wachtel 1978)
<i>Caenorhabditis Elegans (worm)</i>	0.3 and 0.75	TEM cell. Exposure of whole organism.	2-16 h	Unexposed control (exposure not measured) and shielded control.	no	21-27 dBm input power	Differences in stress responses for some exposure times at 0.75 GHz, not for others. Effect was more pronounced in specimen closer to source.	(Daniells et al. 1998)
<i>Caenorhabditis Elegans (worm)</i>	1	TEM cell. Exposure of whole organism.	2.5 h	Sham control. Also heat shock as control.	yes	0.9 – 3 mW/kg	No consistent changes in RNA gene expression over five repetitions of the experiment. Number of significant changes was lower than the expected number of false positives. There were changes for the heat shock group.	(Dawe et al. 2009)
<i>Caenorhabditis Elegans (worm)</i>	0.75-1	TEM cell. Exposure of whole organism.	20 h	Unexposed control (exposure not measured) and shielded control.	no	1 mW/kg	Exposure induced heat-shock response. Increased growth after exposure in comparison to control. Increased percentage of eggs in comparison to shielded control and heat-shocked control.	(de Pomerai et al. 2002)
<i>Eiseniafetida (earthworm)</i>	0.9	TEM cell. Exposure of whole organism.	2-4 h	Unexposed control. Unclear whether this was sham. Exposure was not measured.	unclear	10-120 V/m	All exposure treatments induced significant genotoxic effects. The authors conclude that the exposure has DNA-damaging capacities.	(Tkalec et al. 2013)
<i>Helix Aspersa (snail)</i>	2.45	Cavity. Exposure of ganglia.	30 or 60 min	Sham	yes	13 mW/g	Ganglia of snail were dissected and exposed. Microwave exposure changes membrane conductance (resistance).	(S. L. Arber and Lin 1985)
<i>Helix Aspersa (snail)</i>	2.45	Cavity. Exposure of ganglia.	30 or 60 min	Sham	yes	7, 13, 14 W/kg	Ganglia of snail were dissected and exposed. Exposure was done at different temperatures (8-28°C). Exposure of snail neurons to sinusoidal RF-EMFs for 60	(Simon L. Arber and Lin 1985)

							min at 12.9 W/kg inhibited spontaneous activity and reduced input resistance at 8°C and 21°C, not at 28°C.	
<i>Helix Pomatia</i> (snail)	1.9	Antenna in anechoic chamber. Exposure of whole organism.	1 h	Sham control. Blind treatment.	yes	48 mW/kg (FDTD), 16 V/m (measured)	Reaction time for retraction from a hot plate was measured before and after (sham) exposure (blind). The exposed snails were less sensitive to thermal pain.	(Nittby et al. 2012)
<i>Lymnea Stagnalis</i> (snail)	0.9	Waveguide. Exposure of ganglia.	< 3 min	Unexposed control. Unclear whether this was sham or not.	unclear	0.5-15 W/kg	Dissected neurons in snails' ganglia showed bursting responses when exposed to pulsed RF-EMFs. There was a threshold found for the effect of 0.5 W/kg for pulsed signals. Differences were found between CW and pulsed signals.	(Bolshakov and Alekseev 1992)

Other invertebrates

Table 8 lists those studies that investigated RF-EMF exposure of invertebrates that are not insects. A series of papers (Simon L. Arber and Lin 1985; S. L. Arber and Lin 1985; Seaman and Wachtel 1978; Wachtel, Seaman, and Joines 1975) investigated neural responses under RF-EMF exposure in neural cells of snails (*Helix Aspersa*) and stretch receptors of sea slugs (*Aplysia Californica*). Increased neural firing under RF-EMF exposure was observed for both types of cells in comparison to sham-exposed of unexposed control and dose relationships and/or threshold values were investigated. (Bolshakov and Alekseev 1992) found bursting responses of the neurons in the ganglia of the snail *Lymnea stagnalis* under exposure to RF EMFs at 900 MHz and investigated a threshold and dose-effect. (Nittby et al. 2012) investigated RF-EMF exposure of the *Helix Pomatia* snail at 1.9 GHz (16 V/m). They use a high-quality study design with sham exposure, shielding in an anechoic chamber, measurements of the RF-EMF exposure, and numerical dosimetry. They exposed a set of snails in an anechoic chamber to RF-EMFs and compared their response to high temperatures on a hot plate before and after exposure. (Nittby et al. 2012) found that the exposed snails were less sensitive to thermal pain than sham-exposed snails. RF-EMF exposure of the worm *Caenorhabditis Elegans* was studied in (Daniells et al. 1998; de Pomerai et al. 2002). They observed a stress response in the exposed animals and (de Pomerai et al. 2002) found increased growth of worms after exposure in comparison to an unexposed control. The exposure of the control was not verified. (Dawe et al. 2009) investigated RF-EMF exposure of the same worm at 1 GHz and compared RNA gene expression with a sham exposed group. No effect of exposure was found, while a heat shock did induce an effect. (Tkalec et al. 2013) investigated RF-EMF exposure of the earthworm *Eiseniafetida* in a TEM cell and studied genotoxic effects in comparison to an unexposed control group (exposure not measured). They conclude that the exposure treatments induced significant genotoxic effects and that the exposure has DNA-damaging capacities.

Studies on invertebrates at RF frequencies below 400 MHz

Other studies (Hadjinicolaou 1931; A. M. Kadoum, Ball, and Nelson 1967; Ahmed M. Kadoum 1969; Ahmed M. Kadoum, Ball, and Stetson 1967; Rai et al. 1972; 1971; 1974; 1975; 1977; Tomanova and Vacha 2016; Vacha, Puzova, and Kviclova 2009) also investigated exposure of invertebrates at under exposure to RF-EMFs, but focused on frequencies in the low MHz range, which are out of scope of this review.

3.1.3. Review of effects on plants and fungi

The literature review in this section resulted in 121 publications on RF-EMF exposure of fungi and plants in the targeted frequency range. Out of these, 13 were identified as review papers, 3 only provided dosimetric results, 8 only provided information on dielectric properties and 28 did not cover plant morphogenesis. This resulted in a set of 69 publications that are discussed in this section. Out of those, 31 focused on dielectric heating using RF-EMFs, 33 were lab or experimental studies, and 5 were environmental or observational studies.

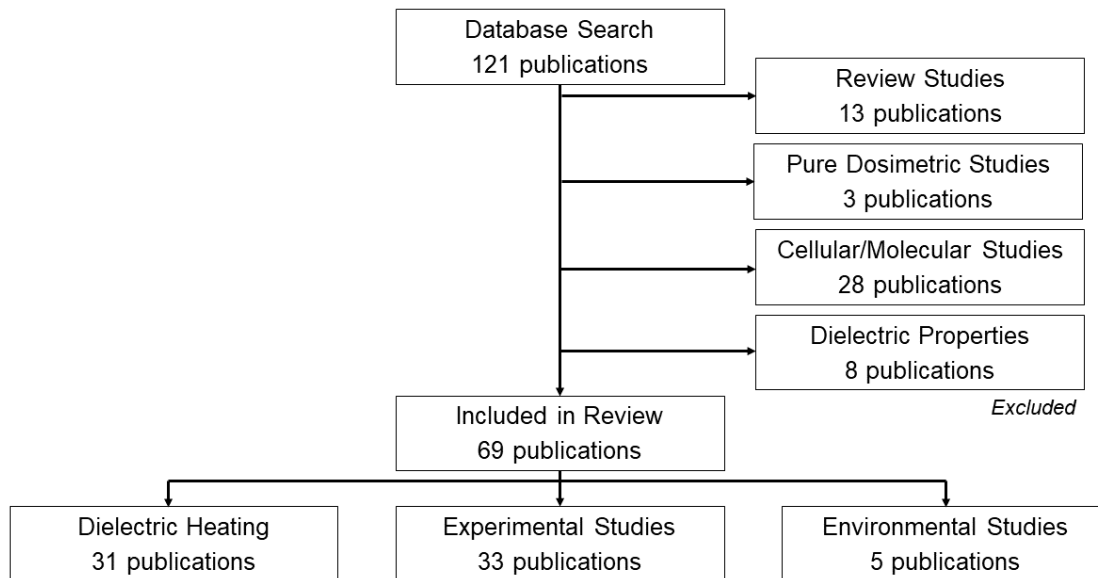


Figure 2 – Flowgraph of the post-processing of the literature review on low-frequency RF-EMF exposure of plants and fungi

Commonly investigated parameters that are used to quantify plant morphogenesis under exposure to RF-EMFs are: length of total plant, epicotyl, hypocotyl, and radicle (root); rhizogenesis (number and length of roots); growth rates of total plant, epicotyl, hypocotyl, and radicle (root); (evolution of) dry and wet mass of the plants (or equivalently water content); and germination rates and speeds. A limited number of papers investigated number and level of abscissions on the plants' stem.

The studies are divided in two parts: those studies that aim to investigate thermal effects of RF-EMF heating and studies that aim to investigate non-thermal effects of RF-EMF exposure or at least do not explicitly aim to heat the investigated plants or fungi.

Table 9 lists studies that investigate heating of fungi and plants using RF-EMFs. All of the references listed in Table 9 demonstrate dielectric heating of plants using RF-EMFs at frequencies lower than 6 GHz. The powers used in those studies listed in Table 9 are much higher than what can be found in the environment.

Several of these studies investigate RF-EMF treatment as a way of pest control in fruits and nuts. Hence, they focus on quality of the fruit and nuts after treatment. Water content is an important aspect of the quality of fruits and nuts. Several studies found reductions in water content after high-power RF-EMF treatment (S. Wang, Tang, et al. 2002; S. Wang et al. 2007a; 2007a; Pande, Mishra, and Singh 2012; Mitcham et al. 2004; Pour-El et al. 1981). However, others did not observe any change in water content (S. Wang et al. 2010; M. C. Lagunas-Solar et al. 2007). (Gao et al. 2010) found no

change in moisture content for unshelled kernels and a reduction for shelled ones. (Karabulut and Baykal 2002) investigated mass loss over time of untreated versus RF-EMF-heated peaches over time and found no difference in mass loss over time.

Several studies investigated germination after RF-EMF heating. Nelson. et al. have investigated germination percentages of a large variety of seeds after high power (>100 kV/m) exposure for very short (several seconds) exposure times. A review on germination percentages of several seeds after RF-EMF exposure to fields between 10 MHz and 2450 MHz was presented in (Stuart O. Nelson and LaVerne E. Stetson 1985). Increased germination percentages were reported for several seed types after specific RF-EMF treatments in terms of time and power. Most of their research effort has been focused on *Medicago Sativa* (alfalfa). In summary, the research presented in (Nelson 1976; S. O. Nelson et al. 1976; Nelson, Kehr, Stetson, and Wolf 1977; Nelson, Kehr, Stetson, Stone, et al. 1977; S. O. Nelson, L. E. Stetson, and W. W. Wolf 1984; Nelson et al. 2002; Iritani and Woodbury 1954) demonstrates drastical increases in germination percentages after short RF-EMF exposure of alfalfa seeds to very high power RF-EMFs. However, these percentages drop drastically after a certain tipping point, increasing the number of dead seeds. A similar behaviour was observed for *Gossypium Hirsutum* (cotton) (Stone et al. 1973). Other studies showed no positive effects on germination, for example (Burk and Nelson 1964) found no improvements in germination of *Nicotiana Tabacum* (Tobacco) after high-power RF-EMF heating. An overview of effects of high-power RF-EMF heating on germination for several plants can be found in Table 9 (peer-reviewed papers with an experimental description) and in (Stuart O. Nelson and LaVerne E. Stetson 1985). More recent studies by other research groups have also investigated germination after RF-EMF heating. (Vadivambal, Jayas, and White 2007) investigated wheat after RF-EMF treatment and observed that germination percentages reduce drastically in comparison with control after treatment and reduce with increasing power (250 to 500 W, treatment < 1 min). (S. Wang et al. 2010) found no effect on germination of *Pisum Sativum*, *Lens Culinaris*, and *Cicer Arietinum* after RF-EMF heating up to 60°C and similar exposure times. Based on the references above and those listed in Table 9, one can conclude that RF-EMF heating will lead to mortality of seeds after a certain exposure time at a certain level. However, some short-durations of RF-EMF heating may lead to higher germination percentages for certain plants.

Finally, it should be mentioned that none of the studies listed in Table 9 use a sham control group, which might be justified by the small exposure times of several minutes or seconds at very high intensities. Additionally, the RF-EMF exposure of the untreated control group is never quantified. However, it is reasonable to assume this exposure was several orders of magnitude smaller than the one of the treated samples.

Table 9 – Effects of RF-EMF (0.01-6 GHz) treatment of plant seeds with the aim of applying dielectric heating

Species	Frequency (GHz)	Exposure conditions	Duration	Exposure level and input	Effect of RF-EMF treatment	Reference
<i>Allium Cepa</i> (onion)	0.01	Parallel-plate electrodes	2 min – 10 min	1 - 2 kV	Onions showed a reduced germination percentage at 1% significance level (5 min exposure), not at other treatment times and amplitudes.	(Iritani and Woodbury 1954)
<i>Cicer Arietinum</i> (Chickpea)	0.027	Parallel-plate electrodes	< 7 min	6 kW	Heating of 3 kg of legumes to 60°C. RF treatments did not significantly affect the moisture content of the three legumes and RF treatment did not affect germination percentages (between 90-100% after RF).	(S. Wang et al. 2010)
<i>Fusarium</i> (fungi)	2.45	Microwave oven	0 - 45 s	800 W	Strong reduction of number of seeds that were contaminated with fungus (< 5% after 30 s).	(Knox et al. 2013)
<i>Gossypium Hirsutum</i> (cotton)	0.04	Unknown	15 - 13 s	1.3-3.1 kV/cm	Heating up to 109°C. Increase of germination up to 25 s, then a decrease.	(Stone et al. 1973)
<i>Glycine Max</i> (soybean)	0.042 and 2.45	Parallel-plate electrodes	< 140 s	0.7 kV/cm	Temperature increase up to 200 °C. Heating is more efficient at 2.45 GHz. Reduction in moisture content with longer treatment.	(Pour-El et al. 1981)
<i>Glycine Max</i> (soybean)	0.043	Dielectric heater	< 2 min	0.65 kV/cm	Heating up to 170°C. Rats that ate the RF heated beans showed faster growth rates, but not faster than positive control (other heating method).	(Borchers et al. 1972)
<i>Glycine Max</i> (soybean)	2.45	Microwave oven	15-60 s	750 W	The exposure reduced seed germination, vigor, and survival of common parasite.	(Reddy et al. 1995)
<i>Juglans Regia</i> (walnut)	0.027	Unknown	< 6 min	12 kW	Heating up to 90°C. Radio frequency treatments reduce the moisture content of walnuts.	(Mitcham et al. 2004)
<i>Juglans Regia</i> (walnut)	0.027 and 0.915	Parallel-plate electrodes	< 10 min (27 MHz), < 16 min (915 MHz)	0.5-0.7 kW/g (27 MHz) and 0.33 kW/g (915 MHz)	Heating up to 70°C. Heating depends on frequency, power level, and configurations.	(Wang, J. Tang, et al. 2003)
<i>Juglans Regia</i> (walnut)	0.027	Parallel-plate electrodes	< 5 min	1 kW/kg	Heating of walnuts until 55°C. Reduced water content after RF exposure for unshelled nuts (not for shelled ones).	(S. Wang, Tang, et al. 2002)

<i>Juglans Regia</i> (walnut)	0.027	Parallel-plate electrodes	< 10 min	< 25 kW	Heating of walnuts until 60°C. Reduced water content in the walnuts, the shells, and the kernels.	(S. Wang et al. 2007b; 2007a)
<i>Lens Culinaris</i> (lentil)	0.027	Parallel-plate electrodes	< 7 min	6 kW	Heating of 3 kg of legumes to 60°C. RF treatments did not significantly affect the moisture content of the three legumes and RF treatment did not affect germination percentages (between 90-100% after RF).	(S. Wang et al. 2010)
M202 (rice)	3 10 ⁻⁷ , 10 ⁻⁶ , 0.01, 0.02	Parallel-plate cavity	< 5 min	100 W	Heating up to 70°C. No difference in moisture content.	(M. C. Lagunas-Solar et al. 2007)
<i>Medicago Sativa</i> (alfalfa),	0.01	Parallel-plate electrodes	2.5 min	Up to 4.5 kV	Increase in germination percentage of hard seed.	(Iritani and Woodbury 1954)
<i>Medicago Sativa</i> (alfalfa)	0.039	Unknown	< 8 s	2.4 kV/cm	Heating up to 90°C. Germination percentages are increased in comparison to unexposed control.	(Nelson 1976)
<i>Medicago Sativa</i> (alfalfa)	0.039	Unknown	Up to 32 s	2 kV/cm	Heating up to 109°C. Increased germination rate up to 24 s, then decrease. Reduction in hard seed	(S. O. Nelson et al. 1976)
<i>Medicago Sativa</i> (alfalfa)	0.039	Unknown	11 - 36 s	1.7 kV/cm	Increase germination and reduction of hard seeds up to 32 s exposure for normal seedlings, then reduction.	(Nelson, Kehr, Stetson, and Wolf 1977)
<i>Medicago Sativa</i> (alfalfa)	0.039	Unknown	≤ 15 s	2.1 kV/cm	Increase in germination and reduction of hard seeds up to 5 s (8% moisture) and 15 s (3% moisture).	(Nelson, Kehr, Stetson, Stone, et al. 1977)
<i>Medicago Sativa</i> (alfalfa)	0.039	Electrodes	≤ 50 s	< 2.4 kV/cm	8y and 20y after exposure percentages of hard seed have reduced for both exposed and control. For some types, there is still an increase in germination 20y. after the treatment.	(S. O. Nelson, L. E. Stetson, and W. W. Wolf 1984)
<i>Medicago Sativa</i> (alfalfa)	0.039	Dielectric heater	< 28 s	1.6 kV/cm	Germination percentages increase up to 18 s and then decreases. Temperature increase up to 120°C.	(Nelson et al. 2002)
<i>Medicago Scutella</i> (snail medic)	0.039 and 2.45	Unknown	12 s (39 MHz) and 50 s (2.45 GHz)	Unknown	Heating up to 84°C. No change in germination.	(S. O. Nelson et al. 1976)
<i>Medicago Truncatula</i> (barrel medic)	0.039 and 2.45	Unknown	19s (39 MHz)/ 70 s (2.45 GHz)	Unknown	Heating up to 74°C. Increase in germination percentage and reduction in hard seed.	(S. O. Nelson et al. 1976)

<i>Melilotus Officinalis</i> (sweetclover)	0.039	Electrodes	< 30 s	1.2 kV/cm	Heating of seeds. No clear positive effect on germination percentages in normal humidity. Increased germination for dried seeds.	(S. O. Nelson and L. E. Stetson 1982)
<i>Microdochium Nivale</i> (fungi)	2.45	Microwave oven	0 to 45 s	800 W	Strong reduction of number of seeds that were contaminated with fungus (< 5% after 30 s).	(Knox et al. 2013)
<i>Nicotiana Tabacum</i> (Tobacco)	0.039	Unknown	≤ 20 s	< 13 kV/inch	Heating of tobacco seeds up to 270 °F. Reduction in germination percentages with increasing time or RF-EMF exposure.	(Burk and Nelson 1964)
<i>Panax Quinquefozium</i> (American Ginseng)	2.45	Cavity	< 200 h	60 W	Water content of plants was drastically reduced after treatment.	(Ren and Chen 1998)
<i>Phaeosphaeria Nodorum</i> (fungi)	2.45	Microwave oven	0 - 45 s	800 W	Strong reduction of number of seeds that were contaminated with fungus (< 5% after 30 s).	(Knox et al. 2013)
<i>Phaseolus Vulgaris</i> (beans)	0.01	Parallel-plate electrodes	2-10 min	1 - 2 kV	No changes in germination up to 2 kV.	(Iritani and Woodbury 1954)
<i>Pisum Sativum</i> (peas)	0.01	Parallel-plate electrodes	2–10 min	1 - 2 kV	No changes in germination up to 2 kV.	(Iritani and Woodbury 1954)
<i>Pisum Sativum</i> (green pea)	0.027	Parallel-plate electrodes	< 7 min	6 kW	Heating of 3 kg of legumes to 60°C. RF treatments did not significantly affect the moisture content of the three legumes and RF treatment did not affect germination percentages (between 90-100% after RF).	(S. Wang et al. 2010)
<i>Prunus Avium</i> (cherry)	0.915	Mode stirred cavity	< 2 min	5 kW	Cherry pit show differential heating in comparison to fruit surface. Inconclusive results on fruit quality.	(Ikediala et al. 1999)
<i>Prunus Dulcis</i> (almond)	0.027	Parallel-plate electrodes	< 12 min	0.75 kW	RF heats up almonds to 63°C in t < 12 min. No change in moisture content for unshelled kernels, reduction for shelled ones.	(Gao et al. 2010)
<i>Prunus Persica</i> (peach)	2.45	Microwave oven	< 2 min	0.4 kW	Heating of the peaches up to 60°C. No changes in weight loss over time.	(Karabulut and Baykal 2002)
Saprophytes (fungi)	2.45	Microwave oven	0 - 45 s	800 W	Strong reduction of number of seeds that were contaminated with fungus (< 5% after 30 s).	(Knox et al. 2013)

<i>Sequoia Sempervivum</i>	2.45	Cavity	< 3 min	< 0.7 kW	Heating of frozen plants up to 40°C. Reduced recovery rate of the other plants in comparison to other heating methods.	(Halmagyi, Surducan, and Surducan 2017)
<i>Stylosanthes Humilis (Townsville stylo)</i>	0.039 and 2.45	Unknown	18/140s	Unknown	Heating up to 98°C. Increase in germination percentage and reduction in hard seed.	(S. O. Nelson et al. 1976)
<i>Trifolium Hirtum (rose clover)</i>	0.039 and 2.45	Unknown	33/210 s	Unknown	Heating up to 109°C. Increase in germination percentage and reduction in hard seed.	(S. O. Nelson et al. 1976)
<i>Trifolium Subterraneum (subterranean clover)</i>	0.039 and 2.45	Unknown	14/70 s	Unknown	Heating up to 101°C. Increase in germination percentage and reduction in hard seed.	(S. O. Nelson et al. 1976)
<i>Trifolium Pretense (red clover)</i>	0.01	Parallel-plate electrodes	2.5 min	Up to 4.5 kV	Increase in germination percentage of hard seed.	(Iritani and Woodbury 1954)
<i>Triticum (wheat)</i>	2.45	Industrial microwave dryer	28 and 56 s	250, 300, 400, and 500 W	No changes in quality of wheat after treatment. Germination percentages reduce drastically in comparison with control and with increasing power.	(Vadivambal, Jayas, and White 2007)
<i>Triticum Aestivum (wheat)</i>	2.45	Microwave oven	0 to 45 s	800 W	Strong reduction of number of seeds that were contaminated with fungi (< 5% after 30 s). Germination of seeds is not reduced for exposure < 15s, but reduced for exposure > 20 s.	(Knox et al. 2013)
<i>Triticum Aestivum (wheat)</i>	2.45	Microwave oven	30 min	0.3, 0.4, 0.5 or 0.6 W/g	Reduction in germination percentages. Reduction in seed vigor. Reduction of infection with fungus <i>Fusarium Graminearum</i> .	(Reddy et al. 1998, 1)
<i>Vigna Radiata</i>	2.45	Microwave oven	40-80 s	180-900 W	Moisture content depends on power and treatment time.	(Pande, Mishra, and Singh 2012)

Three general problems are identified in the lab studies that investigate RF-EMF effects on plants and fungi other than dielectric heating, see Table 10: (1) the quality of control and sham control groups, (2) quantification and stability of the RF-EMFs exposure, and (3) interference between effects due to RF-EMF exposure and other agents (thermal effects and ELF-EMF exposure).

Many of the studied plants need exposure to EMFs (light) in order to develop those parameters that are investigated. This presents a particular issue in RF-EMF exposure experiments with plants with regard to control groups. It is difficult to shield plants from environmental RF-EMFs and keep them exposed to natural light.

No references were found with an unexposed control group (no exposure to RF EMFs in the studied frequency range during the entire experiment). This is a more important issue in this type of studies, since the used RF-EMF levels are close to those that can be found in the environment. Hence, the exposure of the control groups is much closer to the exposure of the exposed groups, than in the studies investigating dielectric heating listed in Table 9. Additionally, the goal of many of these studies is exactly to investigate the effect of exposure to such environmental RF-EMFs, which is difficult without an unexposed control group. (Haggerty 2010) tried studying *Populus Tremuloides* in a faraday cage, but did not quantify exposure in the cage. There are studies in which a control group is shielded from the RF-EMF exposure source that is used in the study, but not from potential other environmental RF-EMF sources. (Magone 1996) shielded their control group during exposure to the studied RF-EMF source, but do not specify whether the plants are shielded further. The shielding was not verified using measurements. (Schmutz et al. 1996; Skiles 2006; Urech, Eicher, and Siegenthaler 1996; Mudalige Don Hiranya Jayasanka Senavirathna et al. 2014) place their control groups in a zone near the RF-EMF source that is either shielded or has a low exposure by design.

Since it is difficult to work with an unexposed control group due to the omnipresence of environmental RF-EMFs, an alternative would be to quantify the exposure of a control group and compare it to an exposed group that has a distinctively different RF-EMF exposure. However, the exposure of the control group is unknown in many studies listed in Table 10. None of the references listed in Table 10 measure RF-EMF exposure of the control group during the entire duration of the experiment. Some references list some instantaneously measured (Schmutz et al. 1996; Mudalige Don Hiranya Jayasanka Senavirathna et al. 2014; Stefi, Margaritis, and Christodoulakis 2017; Skiles 2006; Urech, Eicher, and Siegenthaler 1996; Stefi et al. 2020) or simulated (C. Chen 2014) RF-EMF values for the control group.

As Table 10 shows, many experiments do not use a sham exposed group. The authors of (Tkalec, Malaric, and Pevalek-Kozlina 2005; 2007; Tkalec et al. 2009) state that they have done preliminary experiments, in which '*no significant differences between the growth responses of plants kept in the GTEM cell, but not connected with generator (sham control) and plants outside the GTEM cell were found*'. However, it is not mentioned how long that sham exposure was, while a significant effect of shielding a plant from visible light inside a TEM cell would be expected for certain time durations. Exposure to RF-EMFs of the control group was not measured in (Tkalec, Malaric, and Pevalek-Kozlina 2005; 2007; Tkalec et al. 2009). Hence, it is reasonable to assume that it was different from the RF-EMF exposure in a shielded TEM cell. (Viliche Balint et al. 2016) designed a custom RF-EMF exposure setup in which two identical chambers are used to either generate exposure or sham exposure. However, exposure in the sham chamber is never quantified. Interestingly, in a later study from the same group (Halmagyi, Surducan, and Surducan 2017) on sequoia plants, the authors found differences in shoot length between sham exposure and a control group outside of the exposure setup, after 30 days of sham exposure. Unfortunately, exposure of the sham group and the control group was not quantified in (Halmagyi, Surducan, and Surducan 2017). It thus remains an open question whether the effects observed in this field are caused by placing plants in an exposure setup or whether they are caused by the RF-EMF exposure itself.

Several references listed in Table 10 do not have a control group. In general, the quality of the control groups in this field of research is low. Therefore, potential effects have to be interpreted with this limitation in mind.

A method to overcome the absence of a control or sham-exposed group that is used in some studies, is working with groups of plants with different RF-EMF doses. The goal of such experiments is to show a significant effect for a differential in RF-EMF exposure rather than showing a significant effect in comparison to control. Different approaches are used to generate these different doses: changes in exposure duration (Jinapang et al. 2010; Tkalec, Malaric, and Pevalek-Kozlina 2005; 2007; Y.-P. Chen, Jia, and Han 2009; H. P. Singh et al. 2012; V. P. Sharma and Kumar 2010; Y.-P. Chen, Jia, and Wang 2009; V. P. Sharma et al. 2009; A. Kumar et al. 2016; Ursache et al. 2009; Talei et al. 2013; Tkalec et al. 2009), changes in output power of the RF-EMF source (Tkalec, Malaric, and Pevalek-Kozlina 2005; 2007; Stefi, Margaritis, and Christodoulakis 2017; C. Chen 2014; Jinapang et al. 2010; Grémiaux et al. 2016; Halgamuge, Yak, and Eberhardt 2015; Tkalec et al. 2009), and changes in distance to the RF-EMF source (Oluwajobi, Falusi, and Zubair 2014; Schmutz et al. 1996; Urech, Eicher, and Siegenthaler 1996; Ellingsrud and Johnsson 1993).

The exposure during the actual RF-EMF experiment is quantified or at least estimated in most of the published studies. However, almost none of the studies listed in Table 10 present measurements of RF-EMF exposure of the studied plants before the experiment or during those moments in the experiment when the plants are not in the exposure setup. Some studies list values measured in one or a limited number of time instances (Urech, Eicher, and Siegenthaler 1996; Skiles 2006; Magone 1996; Waldmann-Selsam et al. 2016; Khalafallah and Sallam 2009). It has been shown that environmental RF-EMFs show significant temporal variations (Bolte and Eikelboom 2012; P. Frei et al. 2009; Thielens, Van den Bossche, et al. 2018; Velghe et al. 2019a; Vermeeren et al. 2013), so RF-EMF exposure should ideally be quantified as a function of time during an experiment.

A well-established effect of exposure to RF-EMFs is dielectric heating, see Table 9. Biological material exposed to RF-EMFs will consequently heat up if the RF influx of energy is higher than the outflux of energy. Therefore, thermal effects cannot be excluded in many experiments. (Urech, Eicher, and Siegenthaler 1996) executed an experiment in which two types of lichen were exposed to either RF EMFs at 2.45 GHz or 9.5 MHz. Effects on growth rate were observed for high RF-EMF exposure at 2.45 GHz. However, it was also demonstrated that this exposure leads to a significant increase in temperature, which might explain the changes in growth rate. On the other hand, no effects were observed at 9.5 MHz, a frequency where no efficient thermal heating was expected. Consequently, most of the subsequent studies in the field have implemented temperature control in their experiments. Obviously, exposing plants to high intensities of RF EMFs like those that can be found in microwave ovens will cause significant heating. (Das, Kumar, and Shah 2013a) provide a review of studies that investigate RF-EMF treatment of plants with the aim of controlling pests in the plants. Several of these studies investigate germination and growth rates of plants after exposure to very-high intensity RF-EMFs and find severe reductions in those parameters at high intensities (Das, Kumar, and Shah 2013a). However, such high intensities of RF-EMF levels are extremely uncommon outside of microwave treatment applications.

Table 10 lists the effects on morphogenesis found in this review. This paragraph lists those effects other than dielectric heating that were demonstrated in literature in comparison to a control or sham exposure group, where the RF-EMF exposure of both the exposed group and the control or sham group were measured or quantified at least on one time instance (13 studies, including (Urech, Eicher, and Siegenthaler 1996)). Effects of studies without a control group and without a sham group or without any exposure quantification of the control or sham exposure are not discussed in this paragraph. (Halgamuge, Yak, and Eberhardt 2015) studied *Glycine Max* (soybeans) exposed to 900 MHz in a TEM cell both for short (2 hours at 5.7 or 41 V/m) and long (5 days at 0.57 V/m) exposure. They found effects on the lengths of epicotyl, hypocotyl, or roots, depending on the exposure level and duration in comparison to sham exposure, while no temperature increases were found. (Tkalec,

Malaric, and Pevalek-Kozlina 2005; 2007) studied exposure of *Lemna Minor* (duckweed) at 400 and 900 MHz in a TEM cell at relatively high RF-EMF field strengths (> 10 V/m) for relatively short exposure durations (< 4 h for most conditions, up to 14 h for one condition). They observed some significant effects on growth for some frequencies at specific exposure levels, but no consistent effects over all frequencies and exposure intensities. These effects also depend on number of days after exposure. Water content increased for all exposure conditions at 900 MHz except one. 400 MHz showed some increases in water content and some non-significant differences, depending on the exposure level and duration. The same group investigated root growth in onions (*Allium Cepa*) in the same exposure conditions (Tkalec et al. 2009), but did not observe any consistent effects in growth. (Schmutz et al. 1996) investigated properties of *Piceu Abies* (spruce) and *Fagus Silmticu* (beech) exposed to a horn antenna at 2.45 GHz over a relatively long period of time (3 years). They observed no effect on needle dry weight per branch and no effect on plant height after 3 years of exposure. (Mudalige Don Hiranya Jayasanka Senavirathna et al. 2014) observed reduced small scale growth rates of *Myriophyllum Aquaticum* (parrot feather) under short-term (1 hour) exposure to 2 GHz RF-EMFs. (Stefi, Margaritis, and Christodoulakis 2017) investigated *Zea Mays* (mays) seedlings exposed to a DECT base station operating at 1.8 GHz during 2 weeks at two different levels: medium (0.49 V/m) and high (27 V/m) exposure. Plants with higher RF-EMF exposure were not affected concerning their sprouting potential, biomass production, and leaf structure in comparison to the other group. The same author investigated oleander plants in the same exposure setup (Stefi et al. 2020) and found increased biomass for the exposed plants. (Skiles 2006) investigated *Medicago Sativa* plants that were exposed to RF EMFs emitted a 2.45 GHz horn antenna during 7 weeks and found no significant difference between fresh and dry weights of exposed and control groups. (Bertrand et al. 2018) exposed a culture of yeast to RF-EMFs in a reverberation chamber for a small amount of time (< 1.5 minutes) and found no effect on growth rates. (Viliche Balint et al. 2016) exposed *Phaseolus Vulgaris* beans during eight days to RF-EMF fields at 950 MHz and compared those to a sham control group grown under identical conditions. The exposed group showed increased length, germination, and dry weight. (Ellingsrud and Johnsson 1993) investigated mechanical vibrations in *Codariocalyx Motorius* before and after relatively high RF-EMF exposure and found altered plant rhythms after RF-EMF exposure. However, it is unclear whether this is a thermal effect or not.

A couple of studies investigated plants under environmental RF-EMF exposure. (Balodis et al. 1996) studied growth of *Pinus Silvestris* (pine trees) over a multi-year period in an area where a radar installation was built during the observational period. A negative correlation was observed between the relative additional increment in tree growth and the perceived intensity of the RF-EMF exposure caused by the radar system. However, the paper lacks exposure measurements in particular of the control group. (Magone 1996) studied *Spirodela Polyrhiza* that was grown near a radar installation for a period of 5 days. They observed long-term effects, even on the next generation of plants. Even though the study uses two different types of control groups, they do not present measurements of the control exposure, so it is difficult to attribute any effects to RF-EMF exposure. (Waldmann-Selsam et al. 2016) studied a large set of trees in Germany and did carry out extensive RF-EMF exposure measurements. However, the selection method for the studied trees is questionable and a proper control is not included. The paper does provide an overview of exposure of trees in an urban environment to RF-EMFs. (Haggerty 2010) compared a very limited set of *Populus Tremuloides* grown in shielded, mock-up shielded, and exposed conditions. Finally, (M. Cammaerts and Johansson 2015) studied *Lepidium Sativum* exposed to a functional base station antenna, but did not compare to unexposed plants. All of these studies suffer from low-quality control groups and/or a lack of proper exposure quantification, but they point out interesting options for research on wildlife exposed to RF-EMFs in their natural environment at real exposure levels, something that is very difficult to reproduce in the lab.

Five previous review studies that were targeted specifically on effects of RF-EMF exposure of plants were identified (Alain Vian et al. 2016; Halgamuge 2017; Ribeiro-Oliveira 2019; Czerwiński et al. 2020;

Halgamuge and Davis 2019). These present an overview of studies on plant morphogenesis, but also on gene expressions, potential changes on molecules, or cellular level. There are some review studies on RF-EMF exposure of wildlife that also include plants (Balmori 2009; 2014; Cucurachi et al. 2013; Diprose, Benson, and Willis 1984; Malkemper et al. 2018). This review study did not focus on publications that investigate cellular, molecular, or functional effects in plants of fungi. The database search did result in 28 peer-reviewed publications that investigated these topics (Barsoum and Pickard 1982; Beaubois et al. 2007; Y.-P. Chen 2006; Engelmann 2008; Gustavino et al. 2016; Haider et al. 1994; Jangid et al. 2010; Kouzmanova et al. 2009; Liu, Garber, and Cleary 1982; Qiu et al. 2013; Radic et al. 2007; Rammal et al. 2014; Roux, Vian, et al. 2008; Roux et al. 2006; Sandu et al. 2005; Selga and Selga 1996; M. D. H. J. Senavirathna and Asaeda 2014; Mudalige Don Hiranya Jayasanka Senavirathna, Takashi, and Kimura 2014; Soran et al. 2014; Vela, Wu, and Smith 1976; A. Vian et al. 2006; Alain Vian et al. 2007; Zareh 2015; Roux, Faure, et al. 2008; Qureshi et al. 2017; Chandel et al. 2019a; 2019b; Friedman et al. 2007).

Table 10 – Overview of studies investigating effects of RF-EMF exposure on plant morphogenesis in the lower studied frequency range

Plant Species	Frequency (GHz)	Exposure Conditions	Duration	Control	Sham	Exposure Level	Effect of RF-EMF Exposure	Reference
<i>Allium Cepa</i> (Onion)	0.4 and 0.9	TEM cell	2 or 4 h	Exposure of control is not measured.	unsure	10, 23, 41, 120 V/m	No consistent effect on root growth and some mitotic aberrations were found.	(Tkalec et al. 2009)
<i>Antirrhinum Majus</i> (Snapdragon)	0.2	Dipole	4, 12, 44 h	Unexposed control	no	1.5 V/m (not measured)	Low viability of seedlings developed from exposed flowers (second generation).	(Harte 1975)
<i>Codariocalyx Motorius</i> (Dancing Plant)	0.03	TEM cell	< 400 s	Comparison of rhythm before and after exposure and unexposed control.	no	<0.6 W/cm ²	Effects on leaflet rhythms. Temperature not monitored, so effect might be thermal.	(Ellingsrud and Johnsson 1993)
<i>Daucus Sativus</i> Rohl (carrot)	2.6	Waveguide with pulsed signal.	10 min	Control were non-treated seeds. exposure of control is not measured.	no	241 kV/m	Exposure reduced carrot seed germination.	(Radzevičius et al. 2013)
<i>Fagus Silmticu</i> (beech)	2.45	Horn antenna	3 y and 7 months	Control was exposed to 0.07 W/m ²	yes	1-300 W/m ² (exposed)	No effect on height after 3 years of exposure.	(Schmutz et al. 1996)
<i>Glycine Max</i> (Soy bean)	0.9	TEM cell. CW and GSM modulation.	2 hours	Exposure of control outside of TEM cell was not determined.	Control is sham exposed in TEM cell.	5.7 or 41 V/m	Inhibition of epicotyl (GSM) and root growth (CW) at higher exposure level sham. Effect depends on modulation. At 5.7 V/m only reduced growth of hypocotyl versus sham for CW signal. No temperature increases.	(Halgamuge, Yak, and Eberhardt 2015)
<i>Glycine Max</i> (Soy bean)	0.9	TEM cell. GSM modulation	5 days	Exposure of control outside of TEM cell was not determined.	Control is sham exposed in TEM cell.	0.56 V/m	Reduced growth of epicotyl and hypocotyl was reduced. Root growth was stimulated. No temperature increases were found. Non-parametric testing, 5% sign level.	(Halgamuge, Yak, and Eberhardt 2015)
<i>Glycine Max</i> (Soy bean)	1.8	Growth chamber (100 ×60×50 cm ³)	24 h or 4 h intermittent exposure.	Control in a separate growth chamber with fewer temperature measurements.	no	4.8 or 14.5 W/m ²	Height and fresh weight of soybeans did not differ. Germination differed under RF-EMF treatment. No temperature increases measured in comparison to control.	(C. Chen 2014)

<i>Hibiscus Sabdariffa</i> (Roselle)	n.a.	Resulting field from a GSM base antenna.	30 days	No measurements of RF EMF strength were done for the control group.	no	0.4 up to 1.1 V/m (broadband measurement)	Reduction of flower bud abscission in comparison to control.	(Oluwajobi, Falusi, and Zubair 2014)
Hypogymnia Physodes (lichen/fungi)	2.45 and 0.01	Horn antenna at 2.45 GHz. Dipole antenna at 0.01 GHz.	0,300,550, and 800 days of exposure (2.45 GHz).	Exposure of control was listed.	yes	2, 50, and 500 W/m ² (2.45 GHz). 235 V/m (9.5 MHz)	Substantially reduced growth rate at 500 W/m ² (2.45 GHz). A differentiation between thermal and nonthermal effects was not possible. No effects at 9.5 MHz and also no thermal effects expected.	(Urech, Eicher, and Siegenthaler 1996)
<i>Ipomoea Aquatica</i> (water convolvuluses)	0.425	TEM cell	1 h, 2 h and 4 h	Control was never placed inside the TEM cell. Exposure of control was not monitored.	no	1mW, 100mW, and 10W input power in TEM cell	Growth stimulation of root and total seedling length different at 1 mW and 2 h. power duration level, not at the other levels.	(Jinapang et al. 2010)
<i>Lablab Purpureus</i> (Hyacinth bean)	1.8	Growth chamber (100 x60 x50cm ³)	24 h or 4 h intermittent exposure.	Control in a separate growth chamber with fewer temperature measurements.	no	4.8 or 14.5 W/m ²	Height and fresh weight were reduced with high EMR treatment but not with low treatment (data not included in paper). Germination did not differ under RF-EMF treatment.	(C. Chen 2014)
<i>Lemna Minor</i> (Duckweed)	0.4, 0.9, 1.9	TEM cell.	2 h at 23, 41, and 390 V/m. 4 h at 23 V/m. 14 h at 10 V/m	Exposure of control is not measured.	unsure	10, 23,41, and 390 V/m	Some significant effects on growth for some frequencies at specific exposure levels, but no consistent effects over all frequencies and exposure intensities. Effects also depend on number of days after exposure.	(Tkalec, Malaric, and Pevalek-Kozlina 2005)
Lemma Minor (duckweed)	0.4 and 0.9	TEM cell	2 h or 4 h (only 23 V/m).	Exposure of control is not measured.	unsure	10, 23, 41 and 120 V/m	Water content increases for all exposure conditions at 900 MHz except the 4 hours exposure. 400 MHz showed some increases and some non-significant differences, depending on the exposure level and duration.	(Tkalec, Malaric, and Pevalek-Kozlina 2007)
<i>Lens Culinaris</i>	1.8	Two mobile phones at 2.2 cm on each side of a petri dish. Plants are also exposed to sound	48 h	Control group is exposed to background RF-EMFs and is not exposed to sound from the mobile phones.	no	Authors claim 1mW output power, but this is not verified.	Germination rate was not affected under the specified exposure conditions, but root growth decreased for exposure during dormant phase. However, no details on statistical tests are provided.	(Akbal et al. 2012)
<i>Linum Usitatissimum</i> (flax)	0.9	A mobile phone	2 h	Control were non-treated seeds. exposure of control is not measured.	no	unknown	Increased production of epidermal meristems in the hypocotyl. RF-EMF response is in between control and cold shock.	(Marc Tafforeau et al. 2002)

Medicago Sativa (alfalfa)	2.45	Horn antenna with reflector	7 weeks	Paper claims that exposure of control plants was measured to be zero.	yes	5–12 W/m ²	There is no significant difference between fresh and dry weights between treatment and control	(Skiles 2006)
<i>Moluga bean</i>	1.8	Growth chamber (100 ×60 ×50 cm ³)	24 h or 4 h intermittent exposure.	Control in a separate growth chamber with fewer temperature measurements.	no	4.8 or 14.5 W/m ²	The effect of EMR on the germination rate, fresh weight and height was inconsistent. No temperature increases measured in comparison to control.	(C. Chen 2014)
MR 219 (rice variety)	2.45	Shielded box with dipole antenna.	1, 4, 7, and 10 hours, 6 days	Exposure of control was not measured.	no	1.58 mW input power	10 hours exposure resulted in the highest Germination Percentage and shorter germination time. Root length and shoot length were also increased. There was an increase in temperature in this study that might explain the effect.	(Talei et al. 2013)
Myriophyllum Aquaticum (Parrot feather)	2	In a shielded environment, exposed to patch antenna.	1 h	Exposure not measured over time.	yes	1.42 W/m ²	statistically significant 51 ± 16% reduction in standard deviation of nanometric elongation rate fluctuation (NERF), a parameter that influences growth.	(Mudalige Don Hiranya Jayasanka Senavirathna et al. 2014)
<i>Nerium Oleander</i> (Oleander)	1.9	Base unit of a DECT telephone	2 weeks	Exposure of control is 0.5 V/m.	no	2.85 V/m	Increase in biomass of both the stems and roots of exposed plants.	(Stefi et al. 2020)
Parmeliu Filiucea (lichen/fungi)	2.45 and 0.01	Horn antenna at 2.45 GHz. Dipole antenna at 0.01 GHz.	0,300,550, and 800 days of exposure (2.45 GHz).	Exposure of control was listed.	yes	2, 50, and 500 W/m ² (2.45 GHz). 235 V/m (9.5 MHz)	Substantially reduced growth rate at 500 W/m ² (2.45 GHz). A differentiation between thermal and nonthermal effects was not possible. No effects at 9.5 MHz and also no thermal effects expected.	(Urech, Eicher, and Siegenthaler 1996)
<i>Phaseolus Vulgaris</i> (Red bean)	1.8	Growth chamber (100 ×60 ×50 cm ³)	24 h or 4 h intermittent exposure.	Control in a separate growth chamber with fewer temperature measurements.	no	4.8 or 14.5 W/m ²	Height and fresh weight did not differ with 24 h intermittent exposure, the germination rate was reduced. However, 4 h intermittent exposure did not affect germination rate. No temperature increases measured in comparison to control.	(C. Chen 2014)
<i>Phaseolus Vulgaris</i> (Red Bean)	0.95	Custom designed device.	8 days	Exposure of control is not quantified.	yes	3.8 mW/m ²	Germination rate, the length of stems and roots, and dry matter percentage are higher in exposed group.	(Viliche Balint et al. 2016)
<i>Piceu Abies</i> (spruce)	2.45	Horn antenna	3 y and 7 months	Control was exposed to 0.07 W/m ²	yes	1-300 W/m ² (exposed)	No effect on Needle dry weight per branch length. No effect on height after 3 years of exposure.	(Schmutz et al. 1996)

<i>Pisum Sativum (pea)</i>	Unknown	Two mobile phones were positioned in the 'middle' of a set of seeds.	½ h, 1 h, 2 h, 4 h and 8 h.	No exposure measured of control.	no	Unknow	Germination, length, dry and fresh weight, water content were investigated and authors claim some significant results using ANOVA. However, inspection of the results showed results that are presented as significant but are clearly not significant. Significance level is not mentioned.	(S. Sharma and Parihar 2014)
<i>Rosa Hybrida (Rose bush)</i>	0.9	Mode Stirred Reverberation Chamber	30 min. The exposures were either single (200 V/m) or repeated 3 times, once every 48 h (5 V/m).	Control was not shielded from RF EMF. Exposure of control not measured.	no	5 or 200 V/m	Delayed and reduced growth of secondary axes for low amplitude (5 V/m), not for the high amplitude exposures.	(Grémiaux et al. 2016)
<i>Saccharomyces Cerevisiae (yeast)</i>	0.9 and 2.45	Mode Stirred Reverberation Chamber	94 s	Exposure of control is unknown.	yes	6.1 V/m (0.9 GHz) and 3.44 V/m (2.45 GHz)	No effect on growth	(Bertrand et al. 2018)
<i>Sequoia Sempervivum</i>	2.44	Custom designed device.	≤40 days	Control and sham control. Exposure of control is not quantified.	yes	51 V/m	Increased shoot and root length after 40 days of RF EMF exposure. However, shoot lengths of sham is also different from control.	(Halmagyi, Surducun, and Surducun 2017)
<i>Trigonella Foenum-Graecum (Fenugreek)</i>	unknown	Two mobile phones were positioned in the 'middle' of a set of seeds.	½ h, 1 h, 2 h, 4 h and 8 h.	No exposure measured of control.	no	n.a.	Germination, length, dry and fresh weight, water content were investigated and authors claim some significant results using anova. However, inspection of the results showed results that are presented as significant but are clearly not significant. Significance level is not mentioned.	(S. Sharma and Parihar 2014)
<i>Triticum Gestivum (Wheat)</i>	0.9	Charging cell phone placed in the middle of a set of seeds. ELF-EMF exposure.	72 Hours of exposure.	Exposure not measured and no ELF exposure.	no	n.a.	Authors claim significant reduction in growth, fresh weight, dry weight, and relative water contents. However, test results (anova) are not provided.	(Afzal and Mansoor 2012)
<i>Triticum Gestivum (Wheat)</i>	2.45	Microwave oven	exposed for 0, 5, 10, 15, 20 and 25 s	No exposure measured for control.	n.a.	n.a.	Significant difference in seedlings height and biomass between control group and microwaved groups.	(Y.-P. Chen, Jia, and Han 2009)

<i>Triticum Gestivum (Wheat)</i>	2.45	Microwave oven	exposed for 0, 5, 10, 15, 20 and 25 s	No exposure measured for control.	n.a.	n.a.	Significant difference in seedlings height and root length up to treatment times of 20s. No difference between control and 25 s.	(Y.-P. Chen, Jia, and Wang 2009)
<i>Vigna Radiata (mung bean)</i>	0.9	Shielded chamber (47.5 × 27 × 17.5 cm) with two cell phones.	Exposure times are 0.5 h, 1 h or 2 hours.	Controls were placed in the chamber for the same time as the exposed plants, but without the cell phones. Exposure of control was not measured. Control was not shielded during the entire experiment.	no	5.7 V/m	Rhizogenesis (root number and length) reduced in comparison to control. Significant trend with exposure time. Comparison between exposure times is not possible because plants were not for the same total amount of time in the chamber.	(H. P. Singh et al. 2012)
<i>Vigna Radiata</i>	0.9	a closely shielded chamber (47.5 × 27 × 17.5 cm) with two charging mobile phones. Hence ELF-EMF exposure present	Exposed during 1/2, 1, 2, or 4 h	Controls were placed in another chamber (perhaps not shielded) for an undisclosed amount of time. Exposure of controls was not measured. No ELF-EMF exposure of control.	no	5.7 V/m	Reduction in radicle length, reduction in plumule length, reduction in seedling dry weight. These reductions show a dependency on exposure time. Comparison between exposure times is not possible because plants were not for the same total amount of time in the chamber.	(V. P. Sharma et al. 2010; V. P. Sharma, Singh, and Kohli 2009; V. P. Sharma et al. 2009)
<i>Vigna Radiata</i>	0.9	Cell phone placed in the middle of a set of seeds. ELF exposure.	72 Hours of exposure.	Control is not exposed to ELF-EMFs. Exposure of control is not determined.	no	Unknown	Authors claim significant reduction in growth, fresh weight, dry weight, and relative water contents. However, test results are not provided.	(Afzal and Mansoor 2012)
<i>Vigna Radiata</i>	0.425	TEM cell	1 h, 2 h and 4 h	Control was never placed inside the TEM cell. Exposure of control was not monitored.	no	1 mW, 100 mW, and 10 W input power in TEM cell	Enhanced total seedling length at 100 mW and 1 h exposure.	(Jinapang et al. 2010)
<i>Vigna Radiata</i>	1.8	Growth chamber (100 × 60 × 50 cm ³)	24 h or 4 h intermittent exposure.	Control in a separate growth chamber with fewer temperature measurements.	no	4.8 or 14.5 W/m ²	Reduction of height under some exposure conditions (not all). Germination did not differ under treatment. No change in weight.	(C. Chen 2014)

<i>Zea Mays (mays)</i>	1	TEM cell	1 to 8 h	No control	no	11.5 W input power	Reduced growth of 12-day-old plants. Comparison between exposure times is not possible because plants were not for the same total amount of time in the chamber.	(Răcuci 2015)
<i>Zea Mays (mays)</i>	1.8	Shielded room	½, 1, 2, and 4 h.	Exposure of the control was not measured.	no	332 mW/m ² (measured)	Reduced growth of roots and coleoptiles. Comparison between exposure times is not possible because plants were not for the same total amount of time in the chamber.	(A. Kumar et al. 2016)
<i>Zea Mays (mays)</i>	0.9 and 2.45	TEM cell or microwave oven	24 h (0.9 GHz) or 5 s (2.45 GHz)	Exposure of control group was not determined. Control group was not shielded.	no	900 MHz, 2.2 V/m. 2.45 GHz, 800 W input power	Results on plant length depended on the modulation. The 2.45 GHz short thermal exposure showed no length-of-plant difference to control.	(Răcuci, Miclăuș, and Creangă 2008)
Zea Mays	1.9	Base unit of a DECT telephone	2 weeks	Exposure of control is 0.5 V/m	no	27.5 V/m	Exposed plants were not affected concerning their sprouting potential, biomass, and leaf structure.	(Stefi, Margaritis, and Christodoulakis 2017)
Zea Mays	0.418	TEM cell	1-2-4-12 hours.	no	no	6 W/m ²	No influence on fresh or dry substance.	(Ursache et al. 2009)
Zea Mays	0.935 – 0.960	Base station antenna.	4 weeks	Control plants are grown away from the mobile station. Exposure is unknown.	no	0.7 and 1.5 W/m ²	No change in germination percentage after 8 days. Control has lowest growth rate (cm ² /Week). Changes in leaf thickness were observed.	(Khalafallah and Sallam 2009)

3.2. Higher telecommunication frequencies (6-300 GHz)

3.2.1. Review of effects on vertebrates

The literature review in this section resulted in 140 publications on RF-EMF exposure of vertebrates or cell cultures obtained from vertebrates in the 6-300 GHz frequency range. Out of these, 18 studies only reviewed literature, 6 studies only presented dosimetric results, 5 only reported dielectric properties of vertebrates, 1 only described radar for the detection of vertebrates, and one only presented simulation results. This resulted in 109 studies that investigated effects of high-frequency RF-EMF exposure on vertebrates that were reviewed in this work. Out of those, 29 studies are in vitro, cellular studies and 80 were in vivo studies on animal models. These groups are reviewed separately. Figure 3 shows a flowgraph of the literature review. It should be noted in this section that the literature survey resulted in a significant amount of papers that are published in Russian and these were excluded a priori.

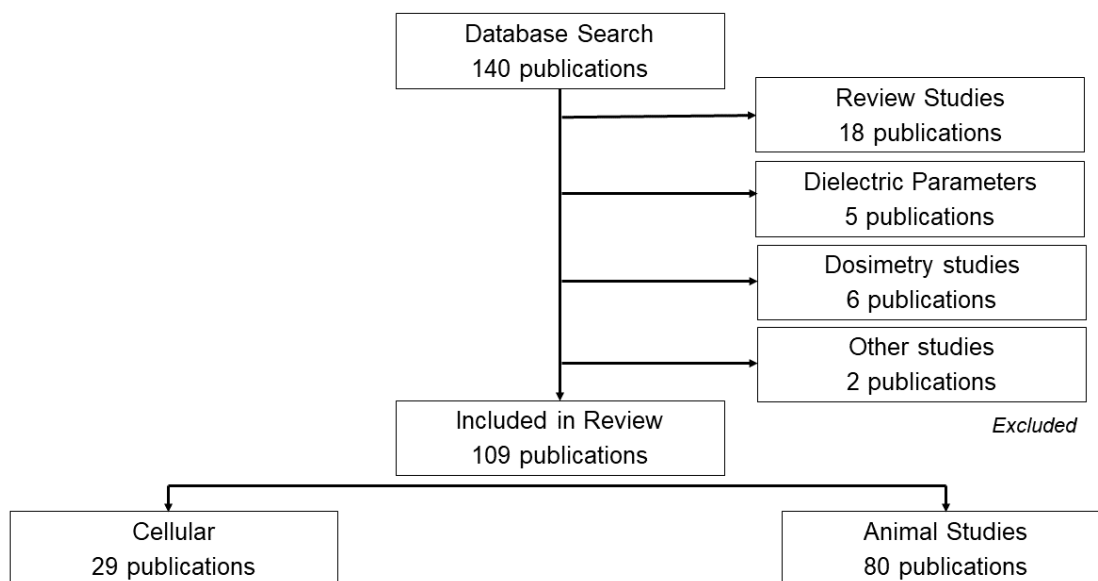


Figure 3 – Flowgraph of the post-processing of the literature review on high-frequency RF-EMF exposure of vertebrates

Dielectric properties of vertebrates in the 6-300 GHz frequency range are presented in (S.I. Alekseev, Gordiienko, and Ziskin 2008; C. Gabriel, Gabriel, and Corthout 1996; S. Gabriel, Lau, and Gabriel 1996a; 1996b; K Sasaki et al. 2015). A series of studies used these dielectric properties or presented their own results on dielectric properties in order to execute dosimetry of vertebrate(s) (cells) in the 6-300 GHz frequency band (Stanislav I. Alekseev and Ziskin 2011; Liberti et al. 2009; Partlow et al. 1981; Kensuke Sasaki et al. 2014; M. Zhadobov et al. 2008; Maxim Zhadobov et al. 2015). (Ning Huansheng et al. 2010) presented results on a radar that focused on the detection of birds and (A B Gapeyev and Chemeris 1999) presented a simulation study of ionic channels. Finally, there have been a series of previous reviews on vertebrates exposed to RF-EMFs in the studied frequency band (Betskii and Lebedeva, n.d.; Brusick et al. 1998; Del Blanco, Romero-Sierra, and Tanner 1973; Gordon et al. 1963; Le Dréan et al. 2013; Obe 2004; A.G. Pakhomov and Murthy 2000; Andrei G. Pakhomov et al. 1998; Ramundo-Orlando 2010; Repacholi 1997; 1998; M. Rojavin 1998; Ryan, D'Andrea, and Jauchem 1999; Tanner and Romero-Sierra 1974; Vaughn 1985; Vecchia 2009; L Verschaeve and Maes 1998; Debouze et al. 2007).

The reviewed studies are split into two main categories: cellular studies (in vitro) and animal studies (in vivo or combined). In the former, a cell line or culture is extracted from a vertebrate and then exposed to RF-EMF fields, while in the latter group the entire organism is exposed to RF-EMFs. Note that in animal studies, it is also possible that cells are extracted after the whole- or partial-body exposure and then processed further in vitro.

Cellular studies

Genotoxicity

The genotoxicity of RF-EMF exposure of vertebrate cell cultures in the 6-300 GHz frequency range was studied in a limited amount of studies. (Garaj-Vrhovac, Horvat, and Koren 1991; 1990) investigated V79 Chinese hamster cells exposed at 7.7 GHz for 15-60 minutes at power densities from 0.5 – 60 mW/cm². They found a dose-dependent reduction in cell survival rates (Garaj-Vrhovac, Horvat, and Koren 1991) and observed significantly higher frequency of specific chromosome aberrations in exposed cells. However, they do not use a sham exposed control and their temperature measurements are limited. (Scarfi et al. 1996) investigated genotoxic effects using cytokinesis-block micronucleus (MN) assay of lymphocyte cells exposed to 9 GHz RF-EMFs at an SAR value of 70 mW/g. They used an unexposed control, combined with a positive control for genotoxicity. The MN frequency increased after RF exposure for both the exposed cells with and without the positive control. They did not use a sham group, but claim to have shown no difference between sham and unexposed control in a previous study.

Neural activation

In vitro cellular studies were used to investigate neural firing and certain action potentials in vertebrate neurons under RF-EMF exposure, see Table 11. (Andrei G. Pakhomov et al. 1997c; 1997a; 1997b) investigated parameters of the compound action potential of frogs' sciatic nerve under pulsed RF-EMF exposure between 40-52 GHz. Using a very-high-quality study design in terms of sham control, they found effects on the action potential that were frequency-specific and cannot be fully explained using thermal effects.

Table 11 – Studies that investigated neural activity in vertebrate cells in vitro under RF EMF exposure between 6 and 300 GHz

Species & cell type	Frequency (GHz)	Exposure Conditions	Duration	control	sham	Exposure Level	Effect of RF-EMF Exposure	Reference
<i>Electroreceptor cells of skates (Rajidae)</i>	37-55	Unclear	< 30 min	Unclear	Unclear	1- 10 mW/cm ²	Transient increase in neural firing rate. It is proposed that this is a thermal effect.	(Akoev, Avelev, and Semenjov 1995)
<i>Frog sciatic nerve</i>	40–52 (pulsed)	Antenna in far-field	10-60 min	Both shielded and sham control. Exposure of sham control was assessed. All equipment was on during sham, but fields were attenuated.	yes	0.24–3.0 mW/cm ²	Changes in parameters of the compound action potential (CAP) under RF-EMF exposure were investigated. At low pulse rates an effect was only found at the highest power densities (2-3 mW/cm ²) and the effect was similar to other heating methods. At high pulse rates a frequency-dependent effect on CAP was observed.	(Andrei G. Pakhomov et al. 1997a)
<i>Frog (Rana Berlandieri or R. Pipiens) sciatic nerve cells</i>	40–52 (pulsed)	Antenna in far-field	38 min	Sham exposed control in shielded chamber	yes	2.5 mW/cm ²	Temporary and reversible decrease of the amplitude and conduction velocity of CAPs. Results depend more in frequency than on intensity.	(Andrei G. Pakhomov et al. 1997b)
<i>Frog (Rana Pipiens) sciatic nerve cells</i>	41 (pulsed)	Antenna in far-field	23 min	Shielded sham control. Exposure of sham control was assessed. All equipment was on during sham, but fields were attenuated.	yes	0.02-2.6 mW/cm ²	High-rate stimulation decreased the CAP, in line with the other studies by the same group. They proposed that the effect is non-thermal.	(Andrei G. Pakhomov et al. 1997c)
<i>Brain slices of Male Sprague-Dawley rats</i>	9.3 (pulsed)	Exposure chamber with open-ended waveguide	2 min	Shielded sham control.	yes	1.57 MV/m (high power)	Population spikes (PS) were evoked by pulsed exposure. The authors reported a transient and fully reversible decrease in the PS amplitude, which was thermal in nature.	(Andrei G. Pakhomov et al. 2003)
<i>Brain slices of neonatal P13–P16 Sprague-Dawley rats</i>	60	Open-ended waveguide	1 min	Unexposed control. Different exposure levels are used.	no	90 mW/cm ² incident, 30 - 800 nW/cm ² at the sample (calculated)	Reversible changes in neuronal firing rate and plasma membrane properties. MMW-induced effects cannot be fully attributed to heating, but heating does show a similar effect. Exposure did increase the temperature.	(Pikov et al. 2010; Siegel and Pikov 2010)

(Andrei G. Pakhomov et al. 2003) also investigated high-energy RF-EMF pulsed exposure at 9 GHz of cortical slices of the rat brain and found that the exposure induced population spikes, which were thermal in nature. (Pikov et al. 2010; Pikov and Siegel 2011) also investigated neural activity in brain slides of rats under RF-EMF exposure. They used RF-EMF exposure at 60 GHz and present calculations that result in very low exposure ($< \mu\text{W}/\text{cm}^2$) of the investigated slices. They did observe temperature increases, even at these very low exposures, and found reversible changes in neuronal firing rate and plasma membrane properties, which might be thermal in nature. They state that the effects cannot be fully attributed to heating. They did not use a sham exposed group, but worked with different doses. (Akoev, Avelev, and Semjenkov 1995) also failed to use a sham group and provide insufficient information on the exposure conditions.

Cellular transformation

(Akoev et al. 1994) investigated exposure of the spinal ganglia of chick embryo to 54 GHz RF-EMFs. They observed a dose-related increase in growth of neurons up to a certain dose ($100 \text{ W}/\text{cm}^2$). Beyond this intensity, the growth was inhibited. The exposure assessment, dosimetry, and control conditions are unclear in this study. Exposure of chondrocytes of Sprague-Dawley (SD) rats was investigated at 30-40 GHz in (Li et al. 2010; 2012) under exposure conditions that are very unclear and with little information on the control. Changes in induced cell apoptosis and mRNA and protein expressions are listed. Stem cells of SD rats exposed to 30-40 GHz RF-EMFs were studied by (Tong et al. 2009; Wu et al. 2009; 2011). They observed changes in mRNA expressions, protein expressions, and induced cell apoptosis, but again under very unclear exposure conditions with little information on the control. (Stensaas et al. 1981) used an experimental procedure of higher quality to study BHK-2VC13 cells exposed to 42 and 74 GHz for 1 h at levels of 320 or 450 mW/cm^2 . They observed temperature increases and changes in cell morphology above a certain threshold temperature in comparison to sham control. Another high-quality study executed by (Haas, Le Page, Zhadobov, Sauleau, et al. 2016) investigated neurite outgrowth in PC12 cells exposed to 60 GHz RF-EMFs at 10 mW/cm^2 during 24 h. They found no effects of exposure on neurite outgrowth in comparison to sham and heated control. Small, insignificant effects could be explained by temperature increases.

Other in vitro studies

Table 12 lists those in vitro studies of vertebrate cells that do not investigate cell transformation, neural activation, or genotoxicity. A series of papers focused on the production of reactive oxygen species (ROS) in mouse peritoneal neutrophils (A.B. Gapeyev et al. 1997; A.B. Gapeyev et al. 1998; Safronova, Gabdoulkhakova, and Santalov 2002). All the studies found that RF-EMF exposure around 42 GHz increased ROS production in comparison to (sham) control at relatively low SAR and incident field levels. (Sun et al. 2012; Titushkin et al. 2009; Shapiro et al. 2013; Geletyuk et al. 1995) investigated the effect of RF-EMF exposure (42-94 GHz) on parameters of ionic channels in vertebrate cells. They found changes in ionic channels under RF-EMF exposure, but attribute those changes to thermal effects (Titushkin et al. 2009; Shapiro et al. 2013). (Melnick, Rubenstein, and Birenbaum 1982) directly exposed rat-liver mitochondria to RF-EMFs at 35 GHz. They found changes in respiratory control, decreases in levels of Ca^{2+} uptake, and increases in extents of Ca^{2+} efflux. These effects could be countered by cooling, which suggests a thermal effect. (Haas, Le Page, Zhadobov, Boriskin, et al. 2016; Haas et al. 2017) investigated the effect of RF-EMF exposure on cellular metabolism and membrane receptors in PC12 cells under 60 GHz exposure, using a high-quality study design with sham exposures, heat control, and numerical dosimetry. They did not find any effects on the parameters they studied that could not be explained thermally. (Samsonov and Popov 2013) investigated showed that RF-EMF exposure increases the rate of microtubule assembly. However, their control conditions and exposure assessment are unclear. They also state that the effect can be explained thermally.

Table 12 – In vitro studies of vertebrate cells under RF EMF exposure between 6 and 300 GHz

Species & cell type	Frequency (GHz)	Exposure Conditions	Duration	Control	Sham	Exposure Level	Effect of RF-EMF Exposure	Reference
<i>Kidney cells of African green marmoset</i>	42	Dielectric waveguide	20-30 min	Unclear	Unclear	100 $\mu W/cm^2$	Effect of RF-EMF exposure on Ca^{2+} and K^+ ionic channels was investigated. Exposure influences channel activity.	(Geletyuk et al. 1995)
<i>PC12 (rat)</i>	60	Horn antenna in far field	24 h	Positive and heat control were used. Sham control is used.	yes	10 mW/cm ² and SAR < 1 kW/kg (FDTD)	Evaluated if RF-EMF exposures impacts expression of membrane receptors at the protein level. No impact of exposure was found. RF-EMF exposure increased temperature.	(Haas, Le Page, Zhadobov, Boriskin, et al. 2016)
<i>PC12 (rat)</i>	60	Horn antenna in far field	24 h	heat control and sham control were used.	yes	5 mW/cm ²	Assessed the impact of MMW exposure on neuronal metabolism. No significant changes in the studied molecules. Any changes could be explained thermally.	(Haas et al. 2017)
<i>Xenopus spinal cord neurons (frog)</i>	94	Open-ended waveguide	< 50 s	Unclear	Unclear	310 W/m ² per 1 mW input, actual value unclear	RF-EMF exposure increases the rate of microtubule assembly. The effect can be explained thermally.	(Samsonov and Popov 2013)
<i>Rat liver mitochondria</i>	35	Near-field of horn	30 min	Sham control	yes	0.5 and 1 W/cm ²	RF-exposure induced losses of respiratory control, decreases in levels of Ca^{2+} uptake, and increases in extents of Ca^{2+} efflux. These effects could be countered by cooling. Loss of respiratory control did not follow a dose-related curve.	(Melnick, Rubenstein, and Birenbaum 1982)
<i>Xenopus Laevis oocytes</i>	60	Open-ended waveguide	< 4 min	Heating control. Unclear what control was with relation to RF-EMF exposure.	unclear	0.18–6 mW/mm ² (exposed side cells, FDTD)	Temperature increase due to mm-wave exposure. Changes in parameters of ionic channels that are consistent with a thermal mechanism. Increases in the action potential firing rate when exposed.	(Shapiro et al. 2013)
<i>P19 (mouse)</i>	94	Open-ended waveguide	Unknown	Unexposed control and heating control.	no	30-60 mW	Exposure increased calcium spiking.	(Sun et al. 2012)
<i>Mouse embryonic stem cell-derived neuronal cells</i>	94	Open-ended waveguide	45 s	Unexposed control	no	18.6 kW/m ²	Ca^{2+} spiking frequency was investigated under RF-EMF exposure. Spiking frequency increased. The effect seemed thermal. Temperature also increased.	(Titushkin et al. 2009)

<i>Mouse peritoneal neutrophils</i>	42 (CW and pulsed)	Open-ended waveguide	20 min	Sham control	yes	7 nW/cm ² – 150 μW/cm ²	It was investigated whether RF-EMF exposure affected production of reactive oxygen species (ROS) by the neutrophils. They observe a reduction in ROS production and a resonant effect at 41.95 GHz. Modulated fields are also compared to CW exposure and result in a changed ROS production.	(A.B. Gapeyev et al. 1998)
<i>Mouse peritoneal neutrophils</i>	42 (CW)	Near-and far-field of antenna	40 min	Unexposed control	no	10 ⁻⁸ -10 ⁻² W/cm ²	Changes in ROS production are observed in comparison to control. Frequency dependence is different for near and far-field and dependence on intensity is also different in near and far field.	(A.B. Gapeyev et al. 1997)
<i>Mouse peritoneal neutrophils</i>	42 (CW)	Horn antenna in far field.	20 min	Sham control	yes	0.45 W/kg (calculated)	RF-EMF increased ROS production and the effect can be inhibited by priming the cells with a reagent.	(Safronova, Gabdoulkhakova, and Santalov 2002)

Animal studies

Dielectric heating and circulatory failure

A series of studies investigated dielectric heating of vertebrates (rodents) and tried to determine the mechanism (circulatory failure) that leads to death due to RF-EMF heating and the thresholds on power density and exposure duration that lead to death, see Table 13. (Deichmann et al. 1959) executed a study where rats and mice were exposed to very high levels of RF EMFs at 24 GHz. They determined dose responses that lead to death of the animals. (Prausnitz and Sutsskind 1962; Poison et al. 1974) executed similar studies at 7 and 9 GHz. (M. R. Frei, Jauchem, and Heinmets 1989, 89) also investigated 9.3 GHz exposure at lower levels. They exposed rats until a 1°C increase in core temperature was reached and recorded changes in heart rate during exposure. A series of studies from the same group (Melvin R. Frei et al. 1995; Ryan et al. 1996; Ryan, Frei, and Jauchem 1997; Ryan et al. 1997; J. R. Jauchem et al. 1997a; Kains et al. 2000; J. R. Jauchem, Ryan, and Tehrany 2004; James R. Jauchem, Ryan, and Walters 2016) investigated exposure of SD rats at 35 GHz (13 W/kg SAR). In most of their studies, rats were exposed until death and certain parameters of the animals were monitored (heart rate, blood pressure, core temperature, and superficial temperature). The main findings are that the superficial temperature increased much stronger under RF-EMF exposure than the core temperature. Blood pressure (arterial) increased during exposure and then decreases until death. They investigated the influence of several drugs on this effect. Heart rate increased during exposure. The effect does not depend on age. The same group also presents results at 10 GHz and 94 GHz (James R Jauchem, Ryan, and Frei 2000; 1999, 199; Millenbaugh et al. 2006) with similar results of RF exposure on body temperature, heart rate, and blood pressure. They observed dose and frequency dependencies. Finally, it must be noted that the exposure levels used in most of the studies listed in Table 13 are relatively high in comparison to environmental exposure levels (at frequencies below 6 GHz) (Bhatt et al. 2016)

Table 13 – Studies that investigated dielectric heating of rodents in the 6-300 GHz frequency range

Species & cell type	Frequency (GHz)	Exposure Conditions	Duration	control	sham	Exposure Level	Effect of RF-EMF Exposure	Reference
<i>SD rats, C59 mice, bantan chicks</i>	24	Horn antenna in shielded chamber	< 500 min	Heating control with IR.	no	20 W input power (mean). 0.02-0.26 W/cm ² in far field.	Rats died after 15 min of exposure in the near field of the antenna. For the far-field exposure the minimal lethal exposure time increased with separation distance from the antenna (43 min at 8 cm and 480 min at 31 cm). 0.028 W/cm ² produced death of a rat after 140 min of exposure. Dose-relationship was also found for the mice. Minimal lethal exposure time increases with decreasing power density. Rectal temperature of animals increased with exposure time. Effects of microwave heating depend on the location of the body that is exposed. RF penetrated further in the body than IR.	(Deichmann et al. 1959)
<i>Swiss albino mice</i>	9 (pulsed)	Horn antenna	4.5 min/day, 5 days/week (52 weeks)	Sham control.	yes	25 W input power (mean). 0.06 – 0.4 W/cm ² , 0.1 W/cm ² in the chronic exposure experiment.	200 mice were exposed during 52 weeks. Rectal temperature increased by 3°C during exposure. 4.5 min was used because 9 min irradiation at this power density causes 50 % mortality. No changes in body weight in comparison to control, no changes in response to temperature changes, and in parameters of the blood. There was testicular degradation in the exposed group in comparison to control.	(Prausnitz and Sutsskind 1962)
<i>SD rats</i>	7.44	Open ended waveguide	≤300 s	Exposure is compared at different frequencies.	no	0.6-6 W/cm ²	Expected mortality at a certain power density was calculated as function of exposure time. For example: at 6 W/cm ² , 50% of the rats are predicted to die after 17 s of exposure. At 0.6 W/cm ² this is 190 s.	(Poison et al. 1974)
<i>SD rats</i>	9.3 (CW and pulsed)	Antenna	Until 1°C temperature increase was reached	CW is compared with pulsed.	no	30 – 60 mW/cm ²	Rectal temperature was increased from 38.5°C to 39°C. Subcutaneous and tympanic temperatures increases colonic temperature during exposure. Heart rate went up during exposure and went back down after. No changes in blood pressure and respiratory rate.	(M. R. Frei, Jauchem, and Heinmets 1989)

<i>SD rats</i>	35	Horn antenna	Until death	no	no	13 W/kg	Animals were exposed until death and temperature and blood flow were measured on several locations in the body. Subcutaneous temperature increased much more than colonic temperature. Heart rate increased during irradiation. Mean arterial pressure was maintained until 42°C and then decreased until death. Mesenteric vascular resistance increased in the beginning of the irradiation and then decreased until death. Circulatory failure manifested itself at colonic temperatures that are relatively normal, while the skin temperature is increased to very high values.	(Melvin R. Frei et al. 1995, 35)
<i>SD rats</i>	35	Horn antenna	Until death	Exposed group was split in two groups: one with the nitric oxide treatment and one with a placebo.	no	13 W/kg	Animals were exposed until mean arterial pressure (MAP) decreased until a certain value. Then the nitric oxide drug was administered or a placebo was administered. There was no change in post drug delivery survival of the rats.	(Ryan et al. 1996)
<i>SD rats</i>	35	Horn antenna	Until death	Exposed rats were split in three age groups	no	13 W/kg	Animals were exposed until death. No differences were measured between the different age groups.	(Ryan et al. 1997)
<i>SD rats</i>	35	Horn antenna	Until death	Exposed group was split in three groups: two were administered different drug that inhibit nitric oxide, and one just water, before irradiation.	no	13 W/kg	Exposure until MAP decreased until certain level then exposure was stopped. Changes in pressure were different for the exposed control and the animals that received the drugs. No changes in survival post treatment.	(Ryan, Frei, and Jauchem 1997)
<i>SD rats</i>	35	Horn antenna	Until death	Exposed group was split in three groups: two was administered esmolol (2 doses) and the other saline water.	no	13 W/kg	Heart rate increased and blood pressure first increased and then decreased for all groups of animals. Animals that received the drug had a dose-dependent decrease in blood pressure in comparison to exposed control (placebo).	(J. R. Jauchem et al. 1997b)
<i>SD rats</i>	94	Horn antenna	Until death	no	no	75 mW/cm ²	Exposure increased temperature. Colonic temperature increases less than the subcutaneous and exposed side temperatures. Arterial blood pressure initially increased and then decreased until death. The heart rate increased during exposure period. Similar results to exposure at 35 GHz.	(James R Jauchem, Ryan, and Frei 1999, 99)

<i>SD rats</i>	1 & 10	Horn antenna	Until death	3 groups: 1 GHz, 10 GHz, and both	no	12 W/kg	Survival was higher at 10 GHz than in the other two groups. Temperature always increased. During irradiation, blood pressure initially increased and then decreased until death. Heart rate increased during exposure.	(James R Jauchem, Ryan, and Frei 2000)
<i>SD rats</i>	35	Horn antenna	< 60 min	no	no	75 mW/cm ²	Oxidative stress occurred in many organs under RF-EMF exposure.	(Kains et al. 2000)
<i>SD rats</i>	35	Horn antenna	Until death	Several groups with placebo and administered drugs at different doses and on different moments relative to the RF exposure. All groups are exposed.	no	13 W/kg	During RF-EMF exposure heart rate increased. Blood pressure first went up and then went down until death. Temperature increased in all groups. Histamine (H1 and H2) antagonists were administered to two groups of exposed animals. Some effects on MAP during exposure and survival after exposure for some doses of drugs.	(J. R. Jauchem, Ryan, and Tehrani 2004)
<i>SD rats</i>	35 & 94	Horn antenna	Unknown	Control with environmental heating in comparison to dielectric heating	no	75-90 mW/cm ²	Temperature distributions over the body are different for each frequency and for the alternative heating method. Time to reach circulatory failure was smallest at highest dose at highest frequency. The authors conclude that body core heating is the major determinant of induction of death due to temperature increase. According to their analysis, the influence of heating of the outer layers (skin and subcutis) is only relevant after a certain threshold on dose.	(Millenbaugh et al. 2006)
<i>SD rats</i>	35	Horn antenna	< 38 min	Sham control	yes	75 mW/cm ²	Cardiovascular and temperature parameters were continuously recorded. Parameters of the blood consistency changed. No changes in blood electrolytes or liver enzymes. Temperature increased in comparison to sham and blood pressure dropped in comparison to sham.	(James R. Jauchem, Ryan, and Walters 2016)

Behaviour

The idea of using high-frequency RF-EMFs to influence the behaviour of birds was postulated in (Tanner 1966). A study in which the behaviour of chickens exposed to modulated, high power RF-EMFs at 9.3 GHz (X-band) was described qualitatively. The effect of radar in the X-band (8-12 GHz) on the behaviour of migratory birds was studied quantitatively in (Bruderer, Peter, and Steuri 1999; Sheridan et al. 2015). No effect of radar exposure on the trajectory of birds was found in (Bruderer, Peter, and Steuri 1999), while (Sheridan et al. 2015) did observe some effects on the behaviour of *Molothrus Ater*, which were not reproduced in two different seasons. Both studies used a very interesting study design with sham exposure to a radar installation. (Nicholls and Racey 2009) used a similar study design to investigate the effect of X-band radar on the behaviour of bats. They observed a reduction in bat activity when radar was on in comparison to when the radar was off.

Genotoxicity

Genotoxicity of RF EMF exposure in the 6-300 GHz band was studied using a limited amount of animal studies. (Kesari and Behari 2009; Paulraj and Behari 2006) investigated DNA breaks in the rat brain after RF-EMF exposure. (Kesari and Behari 2009) exposed rats at 50 GHz with $0.86 \mu\text{W}/\text{cm}^2$ incident power density during 2 h/day for 45 days. They only used 6 exposed animals versus 6 sham exposed animals and found that chronic exposure to RF-EMFs causes DNA double-strand break and decreases the activity of the studied antioxidant enzymes. (Paulraj and Behari 2006) investigated rats exposed for 2h/day for 35 days at 17 GHz at an incident power density level of $1 \text{ mW}/\text{cm}^2$. They also used a limited set of 6 exposed animals that were compared to sham. They found an increase in DNA single strand breaks in brain cells of exposed animals in comparison to sham. (Logani et al. 2004) executed a larger study with 48 BALB/c mice exposed 30 min/day for 3 days to 42 GHz RF-EMFs with a power density of $32 \text{ mW}/\text{cm}^2$. They assessed a potential genotoxicity through the incidence of micronuclei in polychromatic erythrocytes of peripheral blood and bone marrow cells. They stated that this incidence was not different for the exposed groups and conclude that there was no evidence for the genotoxicity of 42 GHz RF-EMFs in the peripheral blood and bone marrow cells of mice.

Cancer

Several studies investigated the effect of RF-EMF exposure on cancer/tumor development. (Ivanov et al. 2005) provide a qualitative description of experiments on carcinogenesis of 37 GHz. The experimental conditions in the study are unclear. The same animals were studied in a high-quality study by (Logani et al. 2006). They exposed the same C57BL/6 mice at 42 GHz at $37 \text{ mW}/\text{cm}^2$ during 30 min using a horn antenna. 50 animals in total were studied in 5 groups. All animals were injected melanoma cells. They used a drug that increased tumor metastases in one group. This was significantly reduced when animals that received that drug were exposed. Millimeter waves also increased natural killer cell activity. (Mason 2001) studied Sencar mice exposed to 94 GHz under two conditions: high exposure ($1 \text{ W}/\text{cm}^2$) during one second and repeated lower exposure ($0.333 \text{ W}/\text{cm}^2$) for 10 s per week. Unexposed and sham controls were used alongside an infrared exposed group. RF-EMF exposure had no effect on tumor incidence and multiplicity. Skin temperature increased $4-5^\circ\text{C}$ in low and $13-15^\circ\text{C}$ in high exposure conditions. (A.A. Radziewsky et al. 2004) investigated the growth of B16 F10 subcutaneous melanoma in mice, which were exposed five times per day for 15 minutes to $13 \text{ mW}/\text{cm}^2$ at 61 GHz. They observed that five daily exposures, if applied starting at the **fifth** day following B16 melanoma cell injection, suppressed tumor growth. The same treatment, started at 1 or 10 days after injection, did not have an effect. Finally, (Rocher et al. 2000) investigated survival of DBA2 mice which were injected leukemia or tumor cells under 60 GHz exposure at $0.5 \text{ mW}/\text{cm}^2$ in comparison to an unexposed control (not a sham exposed control). They observed that the survival of mice with leukemia cells was increased. However, the growth of the studied tumor was enhanced. They used a very limited number of animals.

Reproduction

Three studies investigated the influence of RF-EMF exposure in the considered frequency range on male reproductivity of rats. (Akdağ et al. 1999) investigated SD rats exposed to 9.5 GHz RF-EMFs at 2.65 mW/cm² for 1 hour/day for 13-52 days in comparison to sham exposed animals. 40 animals were sham exposed and 40 were exposed to RF-EMFs. They found increases in rectal temperature after RF-EMF exposure in ¾ study groups. Sperm count decreased for the longest exposure category. Percentage of abnormal sperm count, and weights of testis and epididymis for groups with at least 26 days of exposure. It was suggested that the effect is thermal. (Kesari and Behari 2010) executed a very similar study using Wistar rats, but used only 8 exposed animals at 50 GHz. They found some effects on cellular (sperm) development and antioxidant enzymes. (Manikowska et al. 1979) found disturbances in meiosis of 16 exposed BALB/c mice in comparison to 7 unexposed control animals, a relatively small set of animals, induced by exposure to pulsed 9.4 GHz RF-EMFs of 0.1-10 mW/cm². (S. Kumar, Kesari, and Behari 2011) investigated the sperm of six SD rats exposed to 10 GHz RF-EMFs at 0.21 mW/cm² for 2 hour/day for 45 days in comparison to sham exposed animals. The exposure of the sham group was never measured. However, it is plausible to assume that there was no environmental exposure at 10 GHz. They observed an increase in ROS in the exposed sperm cells, a decline in activity of histone kinase, and an increase in apoptosis.

Nervous system

(Kolosova et al. 1996) investigated exposure of 40 Wistar rats to 54 GHz RF-EMFs at 4 mW/cm² for 10 min every three days. They observed accelerated regeneration of nerve fibers in exposed rats and an increase in conduction velocity of the nerves. No changes in compound action potential were observed (these were observed in the cellular studies). (Stanislav I. Alekseev et al. 2009) investigated RF-EMF exposure at 42 GHz (both CW and pulsed) of the hind paw of Swiss Webster mice at levels of 10 to 200 mW/cm² for exposure times shorter than 100 s. They found that RF-EMF exposure increased skin temperature. Exposure at the incident power density ≥ 45 mW/cm² inhibited the spontaneous electrical activity in the sural nerve in the same hind paw. Nerves increased their firing rate after exposure finished (effect only at ≥ 160 mW/cm²). Heat control shows a similar inhibitory effect on neural firing, but not the same transient after exposure. (Sivachenko et al. 2016) investigated neural activity of the spinal trigeminal nucleus of 13 rats exposed to 40 GHz RF-EMFs generated with an unknown exposure device with an input power of 0.01 mW. 10 min exposure reduced spontaneous firing and suppressed response to a parallel neural stimulation.

Immunology

There have been a series of studies that investigated whether RF-EMF exposure triggers immunological responses in the vertebrate body.

([I Detlavs et al. 1994](#); [I Detlavs et al. 1996](#)) investigated the effect of 42-54 GHz RF-EMF exposure of skin wounds in Wistar rats using a power density of 10 mW/cm². They found that continuous wave signals inhibit inflammatory responses in the skin wound, while modulated RF-EMFs do not show such effects. The exposure conditions that are used in these studies are very unclear and temperature was not controlled.

(Korpan, Resch, and Kokoschinegg 1994) investigated both aseptic and infected skin wounds in rabbits, exposed and unexposed to RF-EMFs at 37 GHz. They found that wound healing was aided by RF-EMF exposure, but it was unclear whether they used sham exposure or not.

(Mikhail A. Rojavin, Tsygankov, and Ziskin 1997) investigated the effect of RF-EMF exposure at 61 GHz on mice that were administered cyclophosphamide (CPA), a drug with a toxic effect. RF-EMF exposure reduced the effect of CPA. The same combination of RF-EMF exposure, this time at 42 GHz, and CPA administration was studied in (Logani et al. 2002; Logani, Agelan, and Ziskin 2002). Treatment with CPA reduced leukocyte and bone marrow cell population (immunosuppression)

and blood contents. RF-EMF exposure did not counteract CPA in these studies. However, a series of follow-up studies (V. Makar et al. 2003; V. R. Makar et al. 2005; 2006) did find that RF-EMF exposure, at 42 and 61 GHz, of mice counteracted the effects of CPA. They conclude that this indicates that there is an involvement of both T-cells and natural killer cells in the immunological response to RF-EMF exposure.

(Fesenko et al. 1999; Novoselova et al. 1999) demonstrated that RF-EMF exposure at 8-18 GHz caused an increase of tumor necrosis factor production in certain macrophages and T-lymphocytes, which is also related to an immune response.

(Lysenyuk et al. 2000) investigated the response of mice to an administered acute inflammation in the leg under exposure to RF-EMFs (43 & 61 GHz) versus sham control. They observed a reduction of the mice's licking response on the presence of the inflammation in exposed animals versus sham exposed animals.

(K. V. Lushnikov et al. 2004; Konstantin V. Lushnikov et al. 2005) investigated RF-EMF exposure of mice at 42 GHz and power density of 0.1 mW/cm². They showed that this exposure has an anti-inflammatory effect, which could be compared to the effect of a certain doses of Diclofenac (an anti-inflammatory drug). A combination of both RF-EMF exposure and a dose of diclofenac resulted in an enhanced effect. The same topic was also investigated by (A.B. Gapeyev, Mikhailik, and Chemeris 2008; Andrew B. Gapeyev, Mikhailik, and Chemeris 2009; Andrew B. Gapeyev et al. 2011). They showed that there is a dose- and frequency-related effect of the RF-EMF exposure and investigated whether modulation of the signals had an influence on the anti-inflammatory effect. They also investigated the role of thymic cells in this response.

(Millenbaugh et al. 2008) investigated gene expressions in the skin of SD rats exposed to 35 GHz RF-EMFs. The rats were exposed in the far field of an antenna at 75 mW/cm² until their colonic temperature increased up to 41-42°C. Gene expression in the skin was compared to a sham control group and a control group that was heated using an alternative technique. The study used 87 SD rats. They observed that the RF-EMF exposure induced 'aggregation of neutrophils in vessels, degeneration of stromal cells, and breakdown of collagen' in the dermis. They also found changes in several gene expressions both after 6 hours and 24 h of exposure. The results are in line with 'thermally related stress and injury in skin while triggering repair processes involving inflammation and tissue matrix recovery'.

Table 14 – Studies that investigated in vivo immunologic effects of RF-EMF exposure in the 6-300 GHz range on vertebrates

Species & cell type	Frequency (GHz)	Exposure Conditions	Duration	control	sham	Exposure Level	Effect of RF-EMF Exposure	Reference
<i>Wistar rats</i>	42-54 (CW and pulsed)	Unclear	30 min/day (5 days)	Unexposed control.	unclear	10 mW/cm ²	Inhibition of inflammatory response in induced skin wound.	(Detlavs et al. 1994)
<i>Wistar rats</i>	42-54 (CW and pulsed)	Unclear	30 min/day (5 days)	Unexposed control.	unclear	10 mW/cm ²	Skin wound inflammation was inhibited in animals exposed to unmodulated RF-EMFs (60 animals).	(Detlavs et al. 1996)
<i>Chinchilla rabbits</i>	37	Horn antenna	30 min/day (5 or 7 days)	Unexposed control	no	1 mW/cm ²	Comparison of rabbits with aseptic and infected skin wounds. The wound healing of both aseptic and infected skin wounds was aided with RF-EMF exposure. The healing process was more active and took less time for exposed animals.	(Korpan, Resch, and Kokoschinegg 1994)
<i>BALB/c mice</i>	61	Near-Field of Horn Antenna	20 min/day for 3 days	Sham control	yes	15 mW/cm ²	RF-EMF exposure reduces the toxic effect of the drug cyclophosphamide (CPA) on cellular immunity.	(Mikhail A. Rojavin, Tsygankov, and Ziskin 1997)
<i>NMRI mice</i>	8-18	Unclear	0.5-7 days	Unexposed cage control (same cage, no exposure)	no	1 μ W /cm ² (2–5 mW/kg)	RF-EMF exposure caused increase of Tumor necrosis factor production in certain macrophages and T-lymphocytes.	(Fesenko et al. 1999)
<i>NMRI mice</i>	8-18	Unclear	5 h	Unexposed control and sham exposure.	yes	1 μ W /cm ² (2–5 mW/kg)	RF-EMF exposure induced an increase tumor necrosis factor production in macrophages and T-cells. Increased mitogenic response in T-lymphocytes after RF-EMF exposure	(Novoselova et al. 1999)
<i>Mice</i>	43, 61	Open-ended waveguide	3 or 10 min	Sham exposure	yes	0.1-7 mW/cm ²	RF-EMF exposure improved the condition of animals that were administered an acute inflammation in the leg Their licking reaction was reduced. Frequency- and dose both influenced the outcomes.	(Lysenyuk et al. 2000)
<i>BALB/C mice</i>	42	Open-ended waveguide	30 min/day for 3 days	Sham control	yes	622 W/kg (31 mW/cm ²)	Treatment with CPA reduced leukocyte and bone marrow cell population (immunosuppression). RF-EMF exposure did not counteract CPA.	(Logani et al. 2002)

BALB/C mice	42	Open-ended waveguide	30 min/day for 3 days	Sham control	yes	622 W/kg (31 mW/cm ²)	RF-EMF exposure before or after CPA administration did not reduce the effect of CPA on mouse blood.	(Logani, Agelan, and Ziskin 2002)
BALB/C mice	42	Open-ended waveguide	30 min/day for 3 days	Unexposed control and sham control	yes	31 mW/cm ²	Effect of RF-EMF exposure on T cell activation, proliferation, and effector functions. These are important for T-cell-related immune responses. RF-EMF exposure counters the immunosuppressive effects of CPA and alters the activation and effector functions of certain T-cells.	(V. Makar et al. 2003)
BALB/C mice	42	Open-ended waveguide	30 min/day for 3 days	Unexposed control and sham control	yes	31 mW/cm ²	CPA treatment caused a marked enhancement in natural killer cell activation. Co-exposure to CPA and RF-EMFs changed that response. RF-EMF exposure can up-regulate NK cell functions	(V. R. Makar et al. 2005)
BALB/c mice	61	Near-Field of Horn Antenna	30 min/day for 3 days	Unexposed control and sham control	yes	31 mW/cm ²	RF-EMF exposure caused upregulation in tumor necrosis factor production in the studied macrophages, which was suppressed by CPA. RF-EMF exposure also enhanced activity of T-cells. RF-EMF exposure accelerate recovery process through an immune response related to T-cells.	(V. R. Makar et al. 2006)
NMRI mice	42	Far-field of antenna	20 min	Unexposed or sham control, unclear.	unclear	0.1 mW/cm ²	RF-EMF exposure reduces the severity of inflammation. The exposure also inhibits production of active oxygen forms in neutrophils that are in an inflammation process.	(K. V. Lushnikov et al. 2004)
NMRI mice	42	Far-field of antenna	20 min	Unexposed control	yes	0.1 mW/cm ²	RF-EMF exposure is compared to Diclofenac drugs dosing. The drug caused a dose-dependent anti-inflammatory effect. RF-EMF exposure reduced the induced footpad edema and hyperthermia associated with the inflammation. This effect was comparable to the effect of diclofenac. Combined treatments caused a partially additive effect.	(Konstantin V. Lushnikov et al. 2005)
NMRI mice	38-70	Far-field of antenna	20-120 min	Sham exposure	yes	0.01-0.1 mW/cm ²	42, 52, and 65 GHz exposure reduced the footpad edema and hyperthermia around the inflammation. Other frequencies were less effective. Bell-shaped dependence on exposure duration at 0.1 mW/cm ² and linear dependence at 0.01 mW/cm ² . Combined treatment with diclofenac was partially additive.	(A.B. Gapeyev, Mikhailik, and Chemeris 2008)

<i>NMRI mice</i>	42-43 (modulated)	Far-field of antenna	20 min	Sham exposure	yes	0.1-0.7 mW/cm ²	No changes in anti-inflammatory effect due to modulation. At some frequencies that were ineffective at CW, the modulation did improve the anti-inflammatory effect.	(Andrew B. Gapeyev, Mikhailik, and Chemeris 2009)
<i>NMRI mice</i>	42	Far-field of antenna	20 min	Sham exposure	yes	0.1 mW/cm ²	Exposure to RF-EMFs changed the consistency of thymic cells. This influences the anti-inflammatory response induced by RF-EMF exposure.	(Andrew B. Gapeyev et al. 2011)
<i>Wistar Rats</i>	37-53	Unclear	2 times 40 min	Unexposed and sham control	yes	20 mW input	Changes in natural killer (NK) cell activity and numbers of c-Fos-positive cells were investigated. Painful electric stimulation decreased the number of NK cells and increased c-Fos-positive cells. This effect could be countered by RF-EMF exposure before and after electric stimulation.	(Shanin 2005)
<i>SD rats</i>	35	Far-field of antenna	Until 41-42°C was reached	Sham control and heating control	yes	75 mW/cm ²	Skin of rats was exposed. There were changes in the dermis after exposure and changes in 56 genes at 6 h and 58 genes at 24 h in the exposed group. The authors state that this indicates that prolonged exposure to RF-EMF causes thermally related stress and injury in skin, while triggering repair processes involving inflammation and tissue matrix recovery. 87 SD rats were studied.	(Millenbaugh et al. 2008)
<i>SD rats</i>	35	Horn antenna	Exposure until colonic T = 41°C	Sham control and heated control with other heating method.	yes	75 mW/cm ²	RF-EMF exposure increased colonic temperature of rats through dielectric heating. Both the alternative heating method (environmental heat) and RF-EMF exposure induce the release of macrophage-activating mediators into the plasma of rats.	(Sypniewska et al. 2010)
<i>New Zealand White rabbits</i>	38	Unclear	20 or 40 min/ day for 10 days	Two unexposed controls.	no	10 mW/cm ²	The modified Mankin Score, the chondrocyte apoptosis, and the expression of caspase-3 and MMP-13 were lower in the group that received the highest dose of RF-EMFs in comparison to control.	(Xia et al. 2012)

(Sypniewska et al. 2010) showed that RF-EMF exposure of rats at 35 GHz increased colonic temperature of rats through dielectric heating. This was compared to an alternative heating method. Both exposures induce the release of macrophage-activating mediators into the plasma of rats, which contributed to immune responses.

Hypoalgesia

Hypoalgesia is a term that is used to indicate a decreased sensitivity to painful stimuli. Several studies have investigated whether RF-EMF exposure can have an effect related to hypoalgesia. A review on this topic was executed by (Usichenko et al. 2006). Most of the A1 published papers in this field come from the research group of prof. Ziskin. They have thoroughly investigated hypoalgesic effects of RF-EMF exposure, mainly at 61 GHz, using Swiss Webster mice (Swiss albino mice) (M A Rojavin and Ziskin 1997; Mikhail A. Rojavin et al. 1998; M A Rojavin et al. 2000; Alexander A. Radziewsky et al. 2000; 2001; A. Radziewsky et al. 2004; A.A. Radziewsky et al. 2008; Alexander A Radziewsky et al. 2002). They demonstrated an prolonged duration of anesthesia after RF-EMF exposure during 15 min at 15 mW/cm² (M A Rojavin and Ziskin 1997; M. Rojavin 1998). They also showed that the same exposure could be used to increase the latency of the mice's response to the cold water tail flick test (M A Rojavin et al. 2000) and determined that this effect is maximized when the nose or paws foot sole were exposed in comparison to the back of the mouse (Alexander A. Radziewsky et al. 2000; 2001). They showed that this treatment did not have negative side-effects on colony activity (Alexander A Radziewsky et al. 2002). They demonstrated that a single exposure at 61 GHz (13 mW/cm² for 15 mins) of the nose suppressed chronic nonneuropathic pain and reduced pain sensitivity of acute pain. However, the treatment was ineffective in the model of chronic neuropathic pain (A. Radziewsky et al. 2004) Finally, they showed that 61 GHz is more effective than 42 or 53 GHz (A.A. Radziewsky et al. 2008).

RF-EMF exposure of the eye

Table 15 lists an overview of all studies that investigated RF-EMF exposure of the eye in the 6-300 GHz range. Two animals are studied: Rabbits and Rhesus Monkeys.

(Richardson, Duane, and Hines 1951) investigated rabbit eyes exposed to 10 GHz pulsed RF-EMFs. 21 rabbits were exposed using a waveguide that was fed up to 67 W of input power. 16/21 animals developed opacities in the eyes within 60 days. (Russell L. Carpenter and Ummersen 1968) investigated exposure of the rabbit eye at 8 and 10 GHz and determined that both frequencies can induce cataract. They determined thresholds for the effect in terms of power versus exposure time. (Birenbaum et al. 2016) determined the same thresholds for induction of changes in the rabbit's lens at 5.5 and 6.3 GHz. The same threshold for cataract-related effects was found at 5.5 and 6.3 GHz. (Kues et al. 1999) investigated ten rabbits, whose eyes were exposed to 60 GHz at 10 mW/cm². They did not observe any detectable ocular damage and they are also the only authors that use an actual sham exposure of the contralateral eye, whereas the other references listed in Table 15 use the contralateral eye as an unexposed control. (Masami Kojima et al. 2009) used high powers at 60 GHz to demonstrate that three types of antennas can be used to cause damages to the eyelids or eye globes. They used 40 rabbits in their study. In a first follow-up study using 30 rabbits (M. Kojima et al. 2012), they demonstrated that 40 GHz exposure increases the internal temperature in the eye, using temperature probes implanted in the eye. They used the same technique to show temperature increases between 18 and 40 GHz in (Masami Kojima et al. 2015). In a very extensive study using 130 rabbits (Masami Kojima et al. 2018), they demonstrated a dose-related influence on corneal temperature and dose-related damages to the cornea due to exposure at 40, 75, and 90 GHz with power densities ranging from 10–600 mW/cm².

Exposure of the eyes of rhesus monkeys (*Macaca Mulatta*) to RF-EMFs in the 6-300 GHz range was first investigated by (R. D. McAfee et al. 1979; Robert D. McAfee et al. 1983). They conditioned monkeys to look into an antenna that is emitting at 9.3 GHz. In contrast to other studies that only

expose one eye of the animals, they exposed both eyes of 12 primates and kept a separate unexposed control group (no sham). The animals were exposed up to 40 times (< 1500 mins in total) to 150 or 300 mW/cm² power densities. The authors did not find any ocular effect induced by such exposures. (Kues et al. 1999) investigated 2 rhesus monkeys exposed to 60 GHz at 10mW/cm² and did not find any ocular effects. (Chalfin et al. 2002; Foster et al. 2003) investigated five monkeys exposed to pulsed RF-EMFs at 35 and 94 GHz. They established thresholds for the induction of corneal lesion and demonstrated that the exposure resulted in large temperature increases in the eye. (Parker et al. 2020) executed a study with 16 monkeys (12 exposed and 4 control) at 94 GHz and measured temperature increases of the eye under exposure to 0.5-2 W/cm². They conclude that the thresholds put forward by (Chalfin et al. 2002) are conservative.

Table 15 – Studies that investigate in vivo exposure of the vertebrate eye exposed to RF-EMFs (6-300 GHz)

Species	Frequency (GHz)	Exposure Conditions	Duration	control	sham	Exposure Level	Effect of RF-EMF Exposure	Reference
<i>Rabbit</i>	10 (pulsed)	Open-ended waveguide	< 20 min	One eye exposed and one as control per animal	no	34-67 W (mean)	21 rabbits were exposed. 16 developed opacities in the eyes within 60 days. These occurred inside the cornea and on the anterior segment of the lens, not on the posterior side (where such effects were observed at lower frequencies).	(Richardson, Duane, and Hines 1951)
<i>Rabbit</i>	5.5 (CW and pulsed) & 6.3 & 70	Open-ended waveguide (low frequencies), horn antenna (70 GHz)	< 150 min	no	no	< 1.1 W input	Both pulsed and CW exposure at 5.5 GHz can induce changes in the rabbit's eye lens. Thresholds for power versus exposure time are determined. Same threshold for cataractogenic effects were found at 6.3 GHz than at 5.5 GHz. Only qualitative results for 70 GHz.	(Birenbaum et al. 1969)
<i>Rabbit</i>	8-10	Open-ended waveguide	< 60 min	no	no	0.15-1.1 W input	8 & 10 GHz exposure can induce cataract in the rabbit's eye. Threshold curves of input power versus time are determined.	(Russell L. Carpenter and Ummersen 1968)
<i>Rabbits</i>	60	Horn antenna	8 h (acute) or 4h/day for 5 days	One eye exposed and one as sham control per animal.	yes	10 mW/cm ²	Single or repeated exposure to 60 GHz CW radiation at 10 mW/cm ² does not result in any detectable ocular damage. 10 rabbits.	(Kues et al. 1999)
<i>Rabbit</i>	60	Horn or lens antenna	6 (lens) or 30 (horn) min	One eye exposed and one as control per animal. Also, pre-exposure of same eye.	no	475 (horn) or 1900 (lens) mW/cm ²	40 rabbits. The three used antennas caused varying damages to the eyelids or eye globes.	(Masami Kojima et al. 2009)
<i>Rabbit</i>	40	Lens antenna	10 min	n/a	n/a	400-100 mW/cm ²	30 rabbits. A temperature probe was implanted in the rabbit's eyes. Temperature increases were measured in all exposures, up to 23°C increase.	(M. Kojima et al. 2012)
<i>Rabbit</i>	18-40	Lens antenna	3 min	n/a	n/a	200 mW/cm ²	16 rabbits. A temperature probe was implanted in the rabbit's eyes. Higher frequencies induced higher temperatures. Temperature increased	(Masami Kojima et al. 2015)

							during exposure in all parts of the eye (cornea, lens, vitreous).	
<i>Rabbit</i>	40, 75, 95	Lens antenna	6 min & 30 min (only 75 GHz)	One eye exposed and one as control per animal. Also, pre-exposure of same eye. Control group with IR radiation.	no	10-600 mW/cm ²	130 rabbits. Dose-related effect on corneal temperature. Dose-related damages to the cornea due to exposure.	(Masami Kojima et al. 2018)
<i>Rhesus Monkeys (Macaca mulatta)</i>	9.3 (pulsed)	Horn antenna	< 700 min	Unexposed control and external control	no	150 mW/cm ² (measured)	Monkeys were conditioned to face the RF-EMF source. They were irradiated 30-40 times and followed-up for one year. No effects on the eye were observed. 12 exposed monkeys.	(R. D. McAfee et al. 1979)
<i>Rhesus Monkeys (Macaca mulatta)</i>	9.3 (pulsed)	Horn antenna	< 1500 min	Unexposed control and external control	no	150 - 300 mW/cm ² (measured)	12 exposed monkeys. Monkeys were conditioned to face the RF-EMF source. They were irradiated multiple times, some at different levels of exposure. No effects on the eye were observed.	(Robert D. McAfee et al. 1983)
<i>Rhesus Monkeys (Macaca mulatta)</i>	60	Horn antenna	8 h (acute) or 4h/day for 5 days	One eye exposed and one as sham control per animal.	yes	10 mW/cm ²	Single or repeated exposure to 60 GHz CW radiation at 10 mW/cm ² does not result in any detectable ocular damage. Only 2 monkeys.	(Kues et al. 1999)
<i>Rhesus Monkeys (Macaca mulatta)</i>	35 & 94 (pulsed)	Open-ended waveguide	< 5 s	One eye exposed and one as control per animal	no	< 11 J/cm ² (2-7 W/cm ² for different durations)	Only 5 animals. Thresholds of 7.5 J/cm ² (35 GHz) and 5 J/cm ² (94 GHz) for corneal lesion. Transient changes were observed at these lesions. Endothelial cell count remained unchanged. Temperature increases up to 30° were measured and correspond well with simulation. 20°C increase is put forward as a threshold for ocular effects.	(Chalfin et al. 2002; Foster et al. 2003)
<i>Rhesus Monkeys (Macaca mulatta)</i>	94	Lens antenna	< 5 min	12 exposed and 4 control.	no	0.5- 2 W/cm ²	16 monkeys. Temperature increase due to exposure. Comments on (Chalfin et al. 2002) and plea for less conservative energy density thresholds for ocular effects. Exposure of 20W/cm ² is safe for the structures of the eye if one can blink.	(Parker et al. 2020)

Other studies

(Vorobyov and Khramov 2002) found changes in the EEG spectra of rabbits exposed to RF-EMFs in the 55-75 GHz frequency band in comparison to control. (Narinyan and Ayrapetyan 2017) investigated the effect of RF-EMF exposure on heart muscle hydration of rats. They found that both sham and RF-EMF exposure at 90-160 GHz increased hydration in comparison to unexposed control.

3.2.2. Review of effects on invertebrates

The literature review in this section resulted in 46 publications on RF-EMF exposure of invertebrates in the 6-300 GHz frequency range. Out of these, 11 focused solely on determining dielectric properties of invertebrates, 3 studies only presented dosimetry results, 3 focused on radar detection of insects, 1 used millimeter waves as a scale model for infrared, and 5 studies were review studies. This resulted in 23 studies that investigated effects of high-frequency RF-EMF exposure on invertebrates. Out of those, 3 studies focused on dielectric heating of insects with the aim of killing them, 12 studies focused on development and genetic effects in insects exposed to RF-EMFs, and 8 studies focused on neural activity induced by exposure to high-frequency RF-EMFs. These three groups are reviewed separately. Figure 4 shows a flowgraph of the literature review.

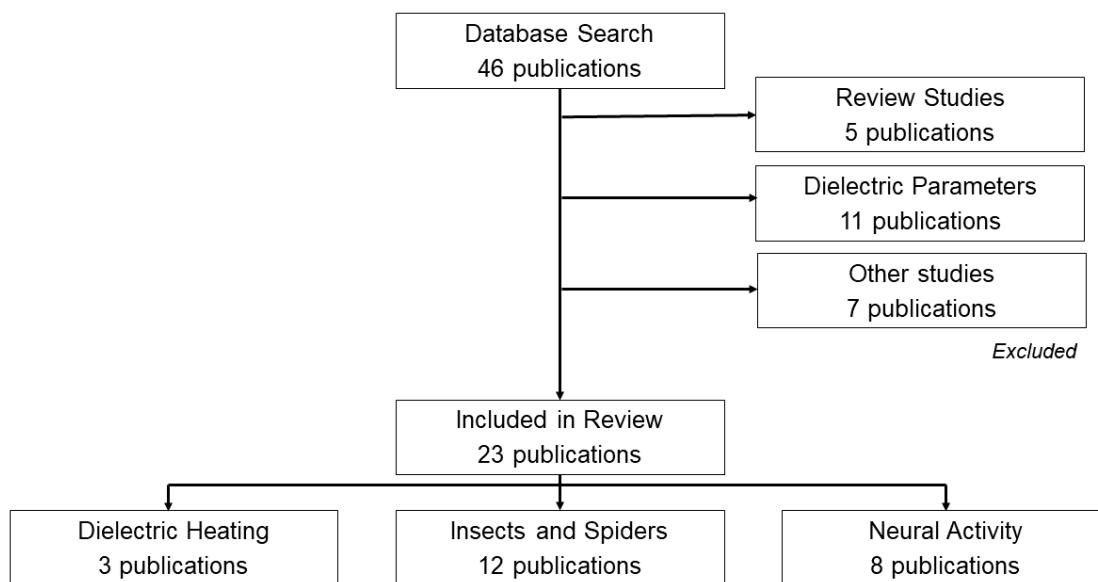


Figure 4 – Flowgraph of the post-processing of the literature review on high-frequency RF-EMF exposure of invertebrates

The literature search resulted in five previous review studies that investigated RF-EMF exposure of invertebrates in the 6-300 GHz range. (Vecchia 2009) investigated a wide range of frequencies and research areas. (Belyaev 1992) focused on potential genetic effects of mm-wave exposure. (Tanner and Romero-Sierra 1974) focused their review on developmental effects, while (Sergii Romanenko et al. 2017) focused on neural activation. Finally, (Del Blanco, Romero-Sierra, and Tanner 1973) provide an overview of the work done in this field prior to 1973.

The dielectric properties of invertebrates, mainly insects, in the 6-300 GHz frequency range were studied up to 70 GHz (Shackelford 1967) in a series of publications (M. Ahmed et al. 2011; Colpitts, Pelletier, and Cogswell 1992; Das, Kumar, and Shah 2013a; Nelson 2004; Nelson et al. 1998; Nelson 2001; Nelson, Bartley, and Lawrence 1997; Nelson 1976; Nelson and L. F. Charity 1972; Shackelford

1967; Hamid, Kashyap, and Cauwenberghe 1968). The main goal of these studies was to determine whether these RF EMFs could be used for pest control in certain stored products. The actual dielectric heating with the aim of exterminating insects is only studied in three publications (Halverson et al. 1996; Watters 1976; Estal et al. 1986). Table 16 shows a summary of their findings. (Halverson et al. 1996) demonstrated dielectric heating of two insects *Sitophilus Zeamais* and *Tribolium Castaneum* at 10.6 GHz at very high powers and short exposure times. This heating caused high mortality rates and temperature increases. (Halverson et al. 1996) also indicate a potential improved differential heating between insects and infested products. (Watters 1976) demonstrated dielectric heating of *Tribolium Confusum* using a lower, but still relatively high power (30 W) with exposure times up to 2 minutes. Temperature increases and mortalities depended on the delivered dose and exposure time. Finally, (Estal et al. 1986) determined dose-mortality relationships for *Ceratitis Capitata* at 9 GHz for different life stages of the insects. The exposures used in the studies listed in Table 16 are relatively high in comparison to environmental exposure to RF-EMFs in the current telecommunication networks at frequencies below 6 GHz (Bhatt et al. 2016; Velghe et al. 2019a; Thielens et al. 2020).

When focusing further on exposure of insects to high-frequency RF-EMFs, (Thielens, Bell, et al. 2018; Thielens et al. 2020) demonstrated that RF-EMFs will be absorbed more efficiently in the body of insects in the 6-300 GHz frequency range than in the lower RF frequency range. However, these studies are limited to numerical dosimetry (and some environmental RF-EMF exposure measurements) and do not present any exposure experiments. Such experiments are executed in the studies listed in Table 17. The discussion of this table is split in three groups of studies: those investigating *Drosophila melanogaster*, *Tenebrio Molitor*, and other insects and spiders.

Table 16 – Overview of studies investigating dielectric heating of insects using high frequency (6-300 GHz) RF-EMF exposure

Species	Frequency (GHz)	Exposure Conditions	Duration	Exposure Level	Effect of RF-EMF Treatment	Reference
<i>Ceratitis Capitata</i> (Mediterranean fruit fly)	9	Waveguide	< 2.25 min	8.6 W/cm ²	A log-linear dose between RF-EMF dose and mortality was determined. The curves depend on the life stage (pupa, adult). High mortalities can be achieved.	(Estal et al. 1986)
<i>Sitophilus Zeamais</i> (Maize Weevil)	10.6	Cavity	< 5 s	9-20 kW	Insect-to-host dissipation ratio of RF power increases at frequencies > 2.45 GHz. Heating up to 64°C. Mortality rates between 53% and 99.9%	(Halverson et al. 1996)
<i>Tribolium Castaneum</i> (Red Flour Beetle)	10.6	Cavity	< 5 s	9-20 kW	Insect-to-host dissipation ratio of RF power increases at frequencies > 2.45 GHz. Heating up to 63°C. Mortality rates between 67% and 99.8%	(Halverson et al. 1996)
<i>Tribolium Confusum</i> (Confused flour beetle)	8.5	Horn Antenna	< 120 s	30 W	Temperature increase from 27°C to 75°C. Mortality depends on exposure time. Highest mortality at 120 s. Mortality higher than 68% after 120 s exposure. Mortality depends on live stage (larva, egg, adult, pupae) of exposure.	(Watters 1976)

Tenebrio Molitor

(Robert L. Carpenter and Livstone 1971) investigated exposure of *Tenebrio Molitor* at 10 GHz at lower input powers than those that are commonly used for dielectric heating, see Table 16. However, they still observed limited dielectric heating. In order to control for the effect of a temperature increase, they worked with a sham-exposed group that was heated using another heating method. In their study, they found higher percentages of deaths and abnormalities in exposed pupae in comparison to an unexposed control, a sham-exposed control, and a sham-exposed control that experienced a temperature increase. From these results, they conclude that the reduction in insect viability must be non-thermal in nature. A follow-up study investigated the same species, exposed to RF-EMFs at 9 GHz (Lindauer et al. 1974). In this study the exposure of the pupae was estimated to be either 8.6 or 17.1 mW/cm². These are relatively high values for incident RF-EMFs and exceed the ICNIRP basic restrictions on RF-EMF exposure in this frequency range (ICNIRP 2020). The study was able to reproduce the results presented by (Robert L. Carpenter and Livstone 1971) and did find higher mortality and higher incidences of abnormalities of RF-EMF exposed groups versus non-exposed control, sham-exposed control, and sham-exposed control heated with an alternative heating mechanism. This study was then again reproduced by (Liu, Rosenbaum, and Pickard 1975), who studied the same insects at the same frequency, for a series of doses and input powers. They confirm higher mortality and higher incidences of abnormalities of RF-EMF exposed groups versus sham-exposed control and show a dose-relationship. They also found significant abnormalities at input powers that are 100 times lower than those used in (Lindauer et al. 1974). However, 0.17 mW/cm² is still a rather high RF-EMF exposure value, but it is lower than the basic restrictions on power density at this frequency (ICNIRP 2020). The authors also executed a parallel dosimetry study (Liu, Rosenbaum, and Pickard 1976). The same insect was then again studied at 9 GHz by (Green, Rosenbaum, and Pickard 1979). They also observed significant abnormalities in adult insects after RF EMF exposure. The authors provide SAR values for the studied insects, but it is unclear how these were obtained. It is unclear what the exposure level was in (Green, Rosenbaum, and Pickard 1979). Given the SAR values (up to 800 W/kg) proved by the authors, the exposure levels were high. To validate whether the effects could be attributed to heating, temperature measurements were. They found that at some non-thermal levels, there were abnormalities observed in the insects. They also found a dose relationship. (Pickard and Olsen 1979) investigated exposure of *Tenebrio Molitor* at 10 GHz and did not find any effects when studying mortality and deformities at 50 W/m².

Drosophila Melanogaster

(Dardalhon, Berteaud, and Averbek 1979) executed an exposure study at 17 and 73 GHz on *Drosophila Melanogaster*. They observed some increases in mortality of exposed eggs, but did not find abnormalities in developed adults after exposure. *Drosophilae* were also studied by (Atli and Ünlü 2006). They exposed larvae and pupae to RF-EMFs at 10 GHz with a field strength of 3.4 V/m for different durations (3-6 hours) and found dose-related increases in the pupation time at non-thermal levels (no decrease in transition percentages). They also observed a reduction in offspring for the group with the longest exposure time during development. The study does not include a sham exposed group and temperature was not monitored, so the effect could be attributed to the experimental setup or the effect could be thermal. (Weisman et al. 2014) investigated the effect of exposure of drosophilae to 100 – 2000 GHz RF-EMFs on lifespan at unknown exposure levels. They observed changes in the mortality of female insects in the second half of their lifespan, but no overall effect on lifespan.

Others

(Koschnitzke et al. 1983) investigated exposure of the glands of *Acricotopus Lucidus* to 64-69 GHz RF-EMFs at exposures up to 6 mW/cm². Certain chromosomes within the glands were analyzed after exposure and an increase in a specific puff was measured in comparison to three different types of control, see Table 17. The web-building capacities of the cross spider *Araneus Diadematus* was

studied under exposure to 9.6 GHz RF-EMFs in (Liddle et al. 1986). No differences were found between webs spun by exposed and sham exposed spiders. (Poh et al. 2017) investigated the behaviour of *Aedes aegypti* mosquitoes in an exposure chamber under RF-EMF exposure between 10 MHz and 20 GHz at an unknown exposure level. They did not observe a difference in behaviour of the mosquitoes in comparison to control, but did not validate whether there was any exposure. (Nicholls and Racey 2009) performed a study in which they investigated the abundance of bats in front of an X-band radar (8-12 GHz). During the same study they placed insect traps in the exposed area and investigated insect abundance during sessions with a fixed radar with two different pulse lengths (0.08 μs and 0.3 μs) in comparison to control sessions where there was no radar signal present. They did not find any differences in insect abundance captured in their traps.

The studies listed in Table 17 are interesting in that sense that they do demonstrate effects, but they do so at rather high RF-EMF levels. It is unclear whether such exposure conditions will occur in the environment, in particular for non-users. It would be extremely interesting to reproduce the studies listed in Table 17 at lower exposure values. Obviously, it is of concern that the literature on (in vivo) invertebrate exposure is limited to 10 publications. Since, most of them found effects and none of them test realistic exposure levels, more research in this field is needed.

Table 18 lists those studies that investigated responses of neural cells exposed to millimeter-waves. These studies dissect certain invertebrates to isolate either a ganglion containing a set of nerve cells (Pikov and Siegel 2011; Sergii Romanenko et al. 2013; 2014; Yamaura and Chichibu 1967), a specific (set of) neurons (S I Alekseev et al. 1997; S I Alekseev and Ziskin 1999), or stretch-receptor organs (Khramov et al. 1991). These are then mounted in front of a waveguide or RF outlet in order to expose them to high-frequency RF EMFs. In parallel the neurons are connected using electrodes in order to register their electrical activity. (Khramov et al. 1991) found a reversible decrease in neural firing of *Astacus Mucus* stretch receptor cells during 34-78 GHz exposure. The exposure values in this study were relatively high and the effects are attributed to be thermal effects. (S I Alekseev and Ziskin 1999; S I Alekseev et al. 1997) investigated activity in the neurons of *Lymnaea stagnalis* under exposure to 60-62 GHz and 75 GHz at SAR values that induced temperature increases up to 2°C. They found alterations in the firing rate of the studied neurons and explain it as a thermal effect.

Table 17 – Overview of studies investigating effects of exposure of insects and spiders to high frequency (6-300 GHz) RF-EMFs

Species	Frequency (GHz)	Exposure Conditions	Duration	Control	Sham	Exposure Level	Effect of RF-EMF Exposure	Reference
<i>Acricotopus Lucidus (Midge)</i>	64-69	Salivary glands are placed in a cavity.	2 h	Two groups: regular sham and sham-control with an alternative heating mechanism. Sham exposed group experienced a 2.5 °C temperature increase. Blinding of study.	yes	< 6 mW/cm ²	Temperature increase was smaller than 0.3°C. Reduction in size of a specific puff of a giant chromosome.	(Koschnitzke et al. 1983)
<i>Aedes Aegypti (Mosquito)</i>	0.01-20	Antenna aimed at exposure chamber	11 h	Unexposed control and shielded control.	yes	Not determined (10 dBm input power)	Movement of mosquitoes was monitored with camera's during exposure. No effect of frequency was found. No clear effect in comparison to control.	(Poh et al. 2017)
<i>Araneus Diadematus (cross spider)</i>	9.6	Anechoic exposure chamber	16 h	Sham control (sham chamber)	yes	0.1 -10 mW/cm ²	The web-spinning ability of the spider was not affected.	(Liddle et al. 1986)
<i>Drosophila Melanogaster (fruit fly)</i>	17 and 73	Horn antenna	2-3 h	Untreated samples	no	100 mW/cm ² (73 GHz) and 60 mW/cm ² (17 GHz)	Some increases in mortality of exposed eggs, number of emerging adults and changes in gender distribution (17 GHz). No consistent effects for exposed larvae and pupae (17 GHz). Change in number of adults and gender distribution (73 GHz). No teratological changes in adults.	(Dardalhon, Berteaud, and Averbeck 1979)
<i>Drosophila Melanogaster (fruit fly)</i>	10	Horn Antenna	3, 4, 5, 6 h	Unexposed control	no	0.0156 W/m ² (measured outside of glass vial, glass partially shields)	No differences in the transition percentages between life stages. Mean pupation time increased with an increasing EMF. Longest exposure time resulted in less offspring.	(Atli and Ünlü 2006)
<i>Drosophila Melanogaster (fruit fly)</i>	100-2000	Unclear	30 min/day	Unexposed control and control in vials	unclear	8.5 mW input power, no exposure quantified	No effect on males, but survival of irradiated females increased in second half of life. Lifespan was not affected.	(Weisman et al. 2014)

<i>Tenebrio Molitor</i> (mealworm beetle)	10	Waveguide	20, 30 or 120 min	3 control groups: unexposed controls, sham-exposed controls, and temperature controls.	yes	80 mW (20 or 30 min) or 20 mW (120 min)	Lower percentage of developed pupae and higher percentages of pupae with abnormal development. In comparison to sham and control. There were temperature increases in the pupae under RF exposure. An alternate heating method was used, which did increase the number of abnormal insects, but not the amount of deaths. The authors thus conclude that the effect must be non-thermal.	(Robert L. Carpenter and Livstone 1971)
<i>Tenebrio Molitor</i> (mealworm beetle)	9	Waveguide	2 h and 4 h	3 control groups: unexposed controls, sham-exposed controls, and temperature controls (heated to 29°C)	yes	8.6 mW/cm ² and 17.1 mW/cm ²	1.5°C temperature increase during exposure. Exposed groups showed significant increased deaths and abnormalities in exposed insects compared to controls. No difference between the exposure groups.	(Lindauer et al. 1974)
<i>Tenebrio Molitor</i> (mealworm beetle)	9	Waveguide	2 h	Sham control	yes	0.05 -20 mW (20 mW ~17 mW/cm ²)	Percentage of normal adults decreases with input power. Percentages of dead and abnormal adults increase with input power. Duration of pupal state increases with power. Significant difference from 0.4 mW/h. Dose is found to be more important than power level.	(Liu, Rosenbaum, and Pickard 1975)
<i>Tenebrio Molitor</i> (mealworm beetle)	9	Waveguide	< 90 min	Sham control	yes	10-320 mW ~ 25 - 800 mW/g	Abnormalities are found in adult insects after RF-EMF exposure at both thermal and non-thermal levels It is shown that relative humidity of the environment also plays a role in the experiment.	(Green, Rosenbaum, and Pickard 1979)
<i>Tenebrio Molitor</i> (beetle)	10	Horn Antenna	4 h	Unclear how the control is performed	Unclear	50 W/m ²	No effect on number of deformities and mortalities in one set. Effect with p=0.051 in one group.	(Pickard and Olsen 1979)
Unknown	8-12	Pulsed Radar	16h	Sham control	yes	18-26 V/m (peak value)	Abundance of trapped insects was unaffected by radar installation being on or off. No difference in abundance of trapped insects as function of pulse length.	(Nicholls and Racey 2009)

Table 18 – Overview of studies investigating neuromodulation due to exposure of invertebrates to high frequency (6-300 GHz) RF-EMFs

Species	Frequency (GHz)	Exposure Conditions	Duration	control	sham	Exposure Level	Effect of RF-EMF Exposure	Reference
<i>Astacus Mucus</i> (Crayfish)	34-78	Stretch-receptor organs were isolated from the animal and placed in front of a dielectric waveguide.	40 s	Control at lower frequency exposure (915 MHz).	no	10 to 215 mW/cm ²	Relative temperature increase < 2°C. Temperature difference increases with increasing power density. Mm-wave exposure causes a decrease in the rate of spontaneous firing. The effect is reversible. No resonant effects are found. Effect at mm-waves is found to be similar to the effect at 915 MHz. A thermal effect is suggested.	(Khramov et al. 1991)
<i>Hirudo Medicinalis</i> (leech)	60	Ganglion was isolated and placed in front of open-ended waveguide.	60 s	no	no	100 to 600 μW/cm ² (different doses)	Reversible changes in the membrane input resistance are observed. These are dose dependent. Temperature was measured and no change was measured. Some effects on neural firing of some neurons.	(Pikov and Siegel 2011)
<i>Hirudo Verbena</i> (leech)	60	Ganglion was isolated and placed in front of open-ended waveguide.	60 s	Control with alternative heating methods (general bath heating and red light)	no	0.9 to 14 mW/cm ² (FDTD dosimetry)	Changes in Neural activity during exposure to mm-wave EMFs. Changes depend on exposure level and are different from other heating methods.	(Sergii Romanenko et al. 2013)
<i>Hirudo Verbena</i> (leech)	60	Ganglion was isolated and placed in front of open-ended waveguide.	60 s	Control with alternative heating methods (general bath heating)		1.0 to 4.0 mW/cm ² (FDTD dosimetry)	Reduction in neural firing rate during exposure. Effect is opposite to the alternative heating method. Also, a narrowing of action potentials.	(Sergii Romanenko et al. 2014)
<i>Lymnaea Stagnalis</i> (pond snail)	60.22–62.22 and 75	Extracted Neuron in a pipet is expose in a solution to an open-ended rectangular waveguide.	< 20 min	Sample placed in the same conditions as the exposed neurons	yes	500–2400 W/kg (in steps) These SARs induced T increases up to 2°C	Millimeter wave irradiation increased the peak amplitudes, activation rates, and inactivation rates of ion currents. The authors conclude that this is a thermal effect.	(S I Alekseev and Ziskin 1999)
<i>Lymnaea Stagnalis</i> (pond snail)	75	Extracted Neuron in a pipet is expose in a solution to an open-ended	< 22 min	no	no	500–4200 W/kg (in steps) These SARs induced T	Alteration of Firing Rate of the studied neuron. The authors suspect that this is a thermal mechanism.	(S I Alekseev et al. 1997)

		rectangular waveguide				increases up to 2.2°C		
<i>Penaeus Japonicas</i> (Kuruma prawn)	11	Extracted ganglion was mounted on the output tip of the mm-wave generator	<10s	Unexposed control	Unclear	$0.5 \times 10^{-4} \text{ W/mm}^3$	Changes in frequencies of neural impulses.	(Yamaura and chichibu 1967)
<i>Procambarus Clarkia</i> (Louisiana crawfish)	11	Extracted ganglion was mounted on the output tip of the mm-wave generator	<10s	Unexposed control	Unclear	$0.5 \times 10^{-4} \text{ W/mm}^3$	Changes in frequencies of neural impulses.	(Yamaura and chichibu 1967)
<i>Richardsonianus Australis</i> (Leech)	60-90	Extracted ganglion of leech was exposed and thermosensitive nociceptor was investigated. Waveguide exposure system.	5 min	Sham exposure and control with other heating method.	yes	100 mW input, 82-170 mW/cm ² in ganglion (simulated), 470 mW/cm ² incident (simulated)	mm-wave irradiation and conductive bath heating activated neurons and increased neural firing. Neuron activation threshold is lower for mm-wave exposure than for conductive heating.	(S. Romanenko et al. 2018)

(Pikov and Siegel 2011) studied exposure of ganglia of *Hirudo Medicinalis* at 60 GHz at lower power densities ($< 10 \text{ W/m}^2$), which induced no temperature changes. Under these exposure conditions, they found reversible, dose-dependent changes in the membrane input resistance and some effects on neural firing of some neurons. (Sergii Romanenko et al. 2014; 2013; S. Romanenko et al. 2018) investigated ganglia of the leeches *Hirudo Verbenae* and *Richardsonianus Australis* exposed at 60-90 GHz and compared their results with neural activity of the ganglia when they are heated using alternative heating methods. They found changes in the neural firing under mm-wave exposure, which were significantly different from control and control heated using other mechanisms. Hence, they conclude that the effect is not thermally induced. (Yamaura and Chichibu 1967) investigated exposure at a lower frequency (10 GHz) of ganglia of *Penaeus Japonica* and *Procambarus Clarkia* at relatively high exposure levels. They found changes in frequencies of neural impulses, but did not provide further insights. From the results presented in Table 18, one can conclude that high-frequency RF exposure can lead to neural responses in invertebrates under in vitro conditions. There are mixed findings in literature whether these effects are thermally induced or non-thermal. Most studies are executed at relatively high exposure levels. It would be interesting to repeat the studies using lower, realistic field intensities.

3.2.3. Review of effects on plants and fungi

The literature review in this section resulted in 54 publications on fungi and plants under exposure to RF-EMFs in the 6-300 GHz frequency range. Out of these, 6 were review papers, 3 were studies that only focused on dielectric parameters, 14 were studies focused on imaging of plants, and 5 were studies that focus on using RF-EMFs for remote sensing. This resulted in 26 studies that investigated effects of high-frequency RF-EMF exposure of fungi and plants. In this section, the 14 studies that focus on fungi (predominantly single-celled yeasts) and the 12 studies that focus on plants (multicellular organisms) are discussed separately. All the studies were lab studies. No environmental studies were found. Figure 5 shows a flowgraph of the literature review.

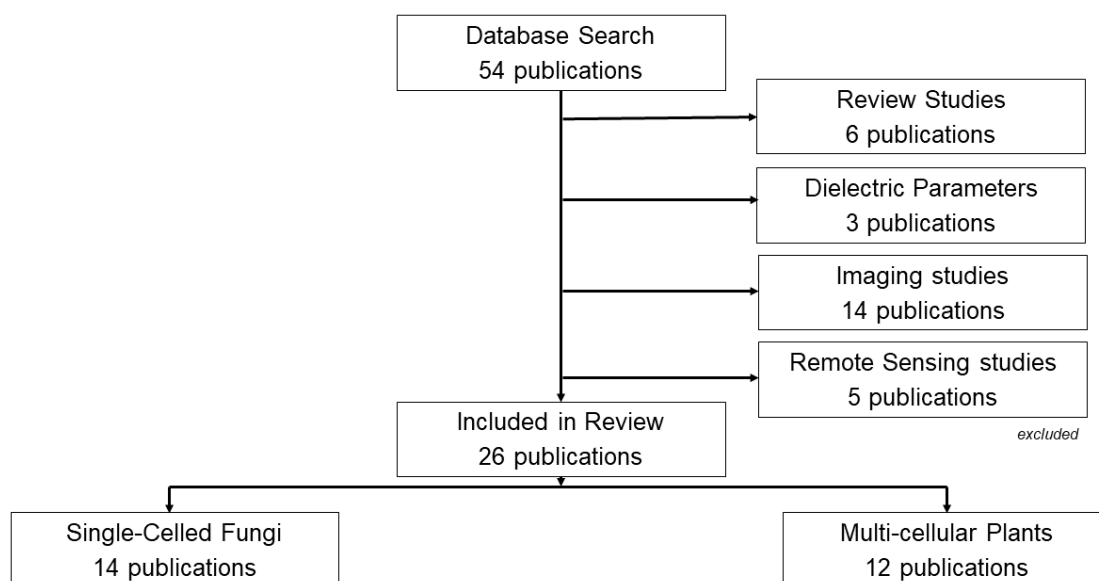


Figure 5 – Flowgraph of the post-processing of the literature review on high-frequency RF-EMF exposure of plants and fungi

Part of the literature that involves plants and fungi in this frequency range focuses on pest management using RF-EMFs. In order to investigate dielectric heating of insects inside of plant

materials, the dielectric parameters of plants are investigated in the 6-300 GHz range in (Nelson 1991; Venkatesh and Raghavan 2004; Das, Kumar, and Shah 2013a). However, these studies do not focus on effects in the plant material after or during exposure.

A set of studies uses RF-EMFs in the higher RF range (> 100 GHz) to perform imaging of plants, so-called THz-imaging. Theoretical papers focus on the reflection and transmission of plant leaves (Afsharinejad et al. 2017; R. Gente et al. 2013; Hadjiloucas, Karatzas, and Bowen 1999), but not on the absorption in the studied frequency range. The technique is applied to several plant species and dependency of reflection and transmission on water content in leaves is demonstrated (Born et al. 2014; Breitenstein et al. 2011; Castro-Camus, Palomar, and Covarrubias 2013; Federici 2012; Ralf Gente and Koch 2015; Jördens et al. 2009; Nie et al. 2017; Santesteban et al. 2015; Song et al. 2018; Torres et al. 2016; Zahid et al. 2019). However, none of the references in this area investigates potential effects of the imaging itself. On the contrary, the technique is proposed as a good candidate for non-destructive imaging because it has no supposed effect on the imaged leaves. Given, the very limited results available in literature on potential effects of such exposures (see Fig. 5) this assumption seems unsupported and more experimental work is necessary to validate the harmlessness of these exposures to plants.

In a related field, the same RF-EMFs are used for remote sensing of growth and water content of plants using satellites (Ferrazzoli and Guerriero 1996; Hunt et al. 2011; Calvet et al. 1994) and mobile (Sawada, Tsutsui, and Koike 2017; Q. Wang et al. 2017) emitters.

Effects of RF-EMF exposure on plants in the 6-300 GHz range were reviewed previously in (Alain Vian et al. 2016), while (Das, Kumar, and Shah 2013a) review dielectric properties and heating of plants in the same frequency range. (M. Tafforeau et al. 2006) reviewed plant responses to environmental stimuli, including RF-EMFs. (Tanner and Romero-Sierra 1974) presented an overview of (mainly unpublished) work done in their lab on plant exposure to high-frequency RF-EMFs. They describe experiments at 10 GHz and intensities up to 190 mW/cm² that found various physiological changes in plants under exposure to such RF-EMFs. Exposures between 10-30 mins at those exposure levels induced wilting in several plants (including *Mimosa Pudica*). The same (unpublished) results were reported in (Del Blanco, Romero-Sierra, and Tanner 1973). No published reproduction of the results was found.

A review paper (Letokhov 1974) reported changes in growth rates of single-celled yeasts at specific frequencies in the 6-300 GHz frequency band. However, the experiments were not described in enough details for scientific reproduction. Nonetheless, there have been a series of studies that investigated growth rates of the yeast *Saccharomyces Cerevisiae*, most commonly at the specific frequency of 42 GHz, see Table 19. Grundler et al. (W Grundler et al. 1982; W. Grundler and Keilmann 1978; 1983; 1989; W. Grundler, Keilmann, and Fröhlich 1977; Werner Grundler et al. 1983) published a series of papers that demonstrated increased growth rates of *Saccharomyces Cerevisiae* after RF-EMF exposure at 42 GHz. However, these papers were contested and others have tried to reproduce (Gandhi 1983) these results, using more strict control (sham) measurements. Both (Furia, Hill, and Gandhi 1986) and (Gos and Eicher 1997) were unable to reproduce the increases in growth rates under RF-EMF exposure and (Jelínek and Šaroch 2007) were unable to observe resonances at 42 GHz in the studied yeast cultures. Survival rates of *Saccharomyces Cerevisiae* after RF-EMF exposure were studied as well, see Table 19. (Dardalhon, Averbek, and Berteaud 1981; 1979) did not find an effect on survival rate of dielectrically heated cells versus cells that were heated using another method. (Pakhomova, Pakhomov, and Akyel 1997) investigated co-exposure to ultraviolet (UV) and 60 GHz RF-EMFs and did not find a change in survival rates after mm-wave exposure. (Dardanoni, Torregrossa, and Zanforlin 1985) studied another single-celled yeast, *Candida Albicans*, exposed to RF-EMFs at 72 GHz. They found changes in growth rate of exposed cells in comparison to sham-exposed cells.

Table 19 – Overview of studies investigating effects of high frequency (6-300 GHz) RF-EMF exposure of *Saccharomyces Cerevisiae*

Frequency (GHz)	Exposure Conditions	Duration	control	sham	Exposure Level	Effect of RF-EMF Exposure	Reference
40-42	Waveguide	4h	Sham exposure with temperature control	yes	20.0 + 0.5 mW.	No statistically significant nonthermal effects. No changes in absorbance in the visible spectrum and growth rate between exposed and sham.	(Furia, Hill, and Gandhi 1986)
9.4, 17, and 70-75	Near field of horn antenna	30 -120 min	Sham exposure	yes	1 to 60 mW/cm ² (70-75 GHz). 1 to 50 mW/cm ² (9-17 GHz)	Dielectric heating is demonstrated. No significant effect on survival in comparison to other heating method.	(Dardalhon, Averbeck, and Bertheaud 1981; 1979)
61.02 and 61.42	Waveguide	30 min	Sham and parallel control	yes	0.13 mW/cm ²	No change in cell survival rate after UV exposure. No change in reverse mutations. Increased incidence of convertants in the RF-EMF-treated cells.	(Pakhomova, Pakhomov, and Akyel 1997)
42	Waveguide and horn antenna	Unclear	Two identical test chambers were constructed in one exposure system to perform concurrent control and test experiments.	yes	0.5 mW/cm ² and 50 mW/cm ²	No effects on cell division rate.	(Gos and Eicher 1997)
42	Waveguide	< 5 h	Non-irradiated control	Unclear	6-34 mW	Increased growth rate in a specific frequency band.	(W Grundler et al. 1982)
42	Waveguide	< 5 h	Non-irradiated control	Unclear	2 mW/cm ²	Increased growth rate.	(W. Grundler and Keilmann 1978)
42	Waveguide and antenna	Unclear	Unclear	Unclear	< 22 mW	Increased growth rate in a specific frequency band.	(W. Grundler and Keilmann 1983)
42	Waveguide and antenna	Unclear	Unclear	Unclear	<30 mW	Increased growth rate in a specific frequency band. No temperature increases.	(Werner Grundler et al. 1983)
42	Waveguide	< 3 h	Non-irradiated control	Unclear	1 mW/cm ²	Increased growth rate in a specific frequency band.	(W. Grundler and Keilmann 1989)

42	Waveguide	Unclear	no	no	A few mW/cm ²	Growth rate stayed constant or enhanced, depending on frequency.	(W. Grundler, Keilmann, and Fröhlich 1977)
42	Resonant cavity	65 min	Unclear	Unclear	Unclear	No emissions of RF-EMF at 42 GHz were measured generating from the yeast cells.	(Jelínek and Šaroch 2007)

Table 20 lists those studies that investigated exposure of multicellular plants to RF-EMFs in the 6-300 GHz frequency range. These studies are faced with the same problems as the studies done on plants and fungi at lower frequencies: (1) the low quality of control groups and absence of sham control groups, (2) quantification and stability of the RF-EMFs exposure. No study was found with an explicitly unexposed control group. However, since the studied RF-EMFs were not widely used at the time of the studies, it is fair to assume that a control group that was not explicitly exposed in an experiment, was actually unexposed. However, only one (Mudalige Don Hiranya Jayasanka Senavirathna, Takashi, and Kimura 2014) of the studied listed in Table 20 has used sham exposure. As discussed in Section 3.1.3, it is currently unclear whether sham exposure for plants in terms of RF-EMF exposure has an effect on the parameters studied in RF-EMF exposure studies. Therefore, it can currently not be assumed whether the results of the studies listed in Table 20 are caused by exposure of the plants to the exposure setup or to the RF-EMFs emitted by the exposure setup. Several studies do not provide statistical test results, see Table 20, and are not discussed further below.

Dielectric heating of plant materials is also possible in the 6-300 GHz range. This was demonstrated by (Watters 1976) for *Triticum Gestivum* at 8.5 GHz. An overview of other studies that investigated dielectric heating in the 6-300 GHz range is provided in (Das, Kumar, and Shah 2013a).

Some non-thermal effects were shown in comparison to control (not in comparison to sham). Exposure of flax to RF-EMFs at 105 GHz during 2 hours was studied in (Marc Tafforeau et al. 2004). They found an increase in the number of meristems in the plant after exposure under non-thermal conditions (temperature did not change). (Scialabba and Tamburello 2002) studied radish seeds exposed to RF-EMFs at 10-13 GHz for a longer period (96 h) at relatively low levels of exposure. They observed reduction in germinations in comparison to control and a dose-response in reduction of germination, reductions in fresh weight in comparison to control, and a dose-related reduction of the hypocotyl length. However, temperature was not measured in (Scialabba and Tamburello 2002). (Bigu-Del-Blanco, Bristow, and Romero-Sierra 1977) investigated exposure of *Zea Mays* to 9 GHz RF-EMFs at 10-30 mW/cm² during 22 - 24 hours of exposure. They found a reduction in growth during the first two weeks of growth. They observed a slight temperature increase of 4°C, but a positive control (no exposure and 4°C) did not show the same effect.

Table 20 – Overview of studies investigating effects of RF-EMF exposure on plants in the higher studied frequency range

Plant Species	Frequency (GHz)	Exposure Conditions	Duration	control	sham	Exposure Level	Effect of RF-EMF Exposure	Reference
<i>Cicer Arietinum</i> (bengal gram),	8.5-10.3	Horn Antenna	30 min, 12 min to 28 min at 9.6 GHz	Control group is a non-exposed group. Exposure of control is not measured.	no	-1 to 5 dBm	No clear effect of frequency. No clear trend with increasing power. Decrease of germination percentage, decrease in root length, decrease in mass %, and reduction of plant height with increase of exposure time. Decrease of germination percentage, decrease in root length, decrease in mass %, and reduction of plant height with increase of exposure time. No statistical test results, significance, or data are shown	(Ragha et al. 2011)
<i>Chara Braunii</i>	6.8-8.2	Microstrip cell	< 4 s	Unclear	Unclear	10 mW/cm ²	No change in Vacuolar resting potential.	(Barsoum and Pickard 1982)
<i>Daucus Sativus</i> Rohl. (carrot)	9.3	Cavity	5, 10, 20 min	Control is unexposed sample. No separate control group. Exposure of control is not measured.	no	606 kV/m	Increased germination and germination energy (only for 5 and 10 mins, not for 20 mins). Decreased height of seedlings.	(Radzevičius et al. 2013)
<i>Lemna Minor</i> (Duckweed)	8	Micro strip antenna	0.5, 1, and 24 h	Control group was placed in identical anechoic chamber.	yes	45–50 V/m	chlorophyll fluorescence parameters changed in comparison to control, but results depend on exposure time at 8 GHz.	(Mudalige Don Hiranya Jayasanka Senavirathna, Takashi, and Kimura 2014)
<i>Linum Usitatissimum</i> (flax)	105	Horn antenna	2 h	Three different type of controls (only Ca, only RF EMF, and both)	Unclear	10 W/m ² (measured)	Increased production of hypocotyl meristems due to 105 GHz exposure under calcium deprivation condition. No temperature increases.	(Marc Tafforeau et al. 2004)
<i>Lycopersicon Esculentum</i> Mill. (tomato),	9.3	Cavity	10 min	Control is unexposed sample. No separate control groups. Exposure of control is not measured.	no	606 kV/m	HPM exposure significantly increased the germination energy and germination for the younger seeds. No effect on the older seeds. Significant positive effect on dry weight and height of tomato seedling shoots	(Radzevičius et al. 2013)
<i>Nitella Flexilis</i>	6.8-8.2	Microstrip cell	< 4s	Unclear	Unclear	10 mW/cm ²	No change in vacuolar resting potential.	(Barsoum and Pickard 1982)
<i>Raphanus Sativus</i> L. (radish),	9.3	Cavity	10 min	Control is unexposed sample. No separate control groups. Exposure of control is not measured.	no	606 kV/m	Higher germination percentage for oldest seeds, not for younger seeds. No clear effect on germination energy. Increased height after RF-EMF exposure.	(Radzevičius et al. 2013)

<i>Raphanus Sativus (Radish)</i>	10.5 and 12.7	Gunn oscillator (open ended cavity) and Horn Antenna	96 h	Control group is a non-exposed group. Exposure of control is not measured.	no	8 or 14 mW and < 0.4 m distance from source. (seem low levels)	Reduction in germinations in comparison to control and reduction of higher dose in comparison to lower dose. Reduction in fresh weight of the highest exposure group. Reduction of the hypocotyl length, with increased reductions for higher powers. Temperature is not measured.	(Scialabba and Tamburello 2002)
<i>Secale Cereale (Rye)</i>	9.2-11.5	Horn antenna	2 h	Control is unexposed sample. No separate control groups. Exposure of control is not measured.	no	0.9 mW/cm2	No significant effect on plant height. Reduction in dry weight for exposed plants (no test results presented).	(Creanga et al. 1995)
<i>Triticum Aestivum (wheat)</i>	8.5-10.3	Horn Antenna	30 min, 12 min to 28 min at 9.6 GHz	Control group is a non-exposed group. Exposure of control is not measured.	no	-1 to 5 dBm	No clear effect of frequency. Plant height, root length and dry weight %, decrease with increasing input power at 9.6 GHz. Decrease of germination percentage and reduction of plant height with increase of exposure time. No statistical test results, significance, or data are shown.	(Ragha et al. 2011)
<i>Triticum Gestivum (wheat)</i>	10.5	Horn Antenna	15, 45, or 75 min.	Control is unexposed sample. No separate control group. Exposure of control is not measured.	no	unknown	14 days after exposure the root and shoot lengths, and fresh mass were increased.	(Hamada 2007)
<i>Triticum Gestivum (wheat),</i>	8.5	Horn antenna	< 120 s	Control is unexposed sample. No separate control groups. Exposure of control is not measured.	no	30 W	Heating of wheat up to 60°C.	(Watters 1976)
<i>Vigna Aconitifolia (moth bean)</i>	8.5-10.3	Horn Antenna	30 min, 12 min to 28 min at 9.6 GHz	Control group is a non-exposed group. Exposure of control is not measured.	no	-1 to 5 dBm	No clear effect of frequency. Plant height, root length and dry weight % increase with RF-EMF power. Decrease of germination percentage, decrease in root length, decrease in mass %, and reduction of plant height with increase of exposure time. No statistical test results, significance, or data are shown	(Ragha et al. 2011)
<i>Vigna Radiata (green gram)</i>	8.5-10.3	Horn Antenna	30 min, 12 min to 28 min at 9.6 GHz	Control group is a non-exposed group. Exposure of control is not measured.	no	-1 to 5 dBm	No clear effect of frequency. No clear trend with increasing power. Decrease of germination percentage, reduction in root length, and reduction of plant height with increase of exposure time. No statistical test results, significance, or data are shown.	(Ragha et al. 2011)
Zea Mays (Mays)	9	Unknown	22-24 h	Control is unexposed. Exposure of control is not measured.	no	10-30mW/cm ² (measured)	Temperature increases up to 4°C at highest exposure level, but positive control for temperature was investigated and had no effect. Reduction in growth during the first two weeks after exposure.	(Bigu-Del-Blanco, Bristow, and Romero-Sierra 1977)

Zea Mays (Mays)	10.75	Exposure to horn antenna	1-2-4-12 hours.	Control is unexposed sample. No separate control groups. Exposure of control is not measured.	no	1 mW/cm ²	Wet and dry mass increased in the exposed plants in comparison to the non-exposed ones. However, these plants were also older plants in a growing phase.	(Ursache et al. 2009)
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4. Limitations

Non-ionizing EMFs are used for other applications than telecommunication. Hence, there can be exposure to RF-EMFs at frequencies that are not included in the studied frequency bands (Bhatt et al. 2016; Velghe et al. 2019a; ECC 2019) (see Table 1). However, these are unrelated to the deployment and operation of fifth generation networks (ECC 2019; Pujol et al. 2020). Therefore, studies that investigate RF-EMF exposure at such frequencies and potential effects of such exposures are not included in this review, see Section 2.2.

In the lower RF-EMF frequency range (< 200 MHz), there have been previous studies that have demonstrated that relatively low intensity RF-EMF exposure, i.e. non-thermal levels of exposure, can disturb magnetoception in organisms (Tomanova and Vacha 2016; Vacha, Puzova, and Kviclova 2009; Engels et al. 2014; Hiscock et al. 2016; 2017; Hore and Mouritsen 2016; Mouritsen 2018; Ritz et al. 2004; Schwarze et al. 2016; Bartos et al. 2019; Granger et al. 2020; Kavokin et al. 2014; Malkemper et al. 2015). However, these frequencies are not used in 5G telecommunication networks and these studies are hence not reviewed in this work.

In order to provide structure to the review, the study is divided in six categories based on frequency of exposure and taxonomy group. Three groups were used: (1) invertebrates, (2) vertebrates, and (3) plants and fungi. By grouping fungi and plants into one category, this review might give the impression that these two types of species are associated to one another, while plants and fungi are two distinct taxonomies with different properties. The grouping of plants and fungi into one category in this review has no biological basis, but is chosen for two other reasons. First, plants and fungi have been grouped in the previous literature reviews in this field (Cucurachi et al. 2013; Malkemper et al. 2018; Balmori 2009). Second, a separate fungi section in the lower studied frequency range would result in a very limited set of papers (1 or 2). Hence, the choice was made to group plants and fungi into one category.

In the review and discussion of exposure outcomes, it was chosen not to provide a hierarchy between the different exposure outcomes, whereas previous reviews in this field have categorized according to exposure outcome and have distinguished between different responses (Vecchia 2009). In this report it is left up to the reader to prioritize between different potential outcomes of RF-EMF exposure.

The meta-review in Section 3.1.1 does not include a discussion on reviews that cover exposure outcomes related to cancer, reproduction, and development, even though such reviews exist. These topics are reviewed in a parallel study by the STOA.

5. Conclusions

5.1. Lower telecommunication frequencies (450 MHz-6 GHz)

5.1.1. Vertebrates

Cellular studies

Out of those review studies that focused on cellular genotoxicity of RF-EMF exposure, five review studies explicitly concluded that the genotoxic effect of RF-EMF exposure at low levels is (very) weak or inexistent. Two review studies concluded that there is a genotoxic effect, but these are based on a very limited selection of the available literature. The other studies, including the most recent and largest review study on cellular genotoxic effects of RF-EMF exposure (Vijayalaxmi and Prihoda 2018) either did not draw any conclusions or state that the available literature shows mixed results or is inconclusive. Reviews on the effect of RF-EMF exposure on cellular transformation and particular on apoptosis presented mixed conclusions. Most reviews did not draw any conclusions. Those that did present a conclusion state that they did not find an effect on RF-induced apoptosis and weak evidence on cellular replication. However, it seems that these conclusions are mainly supported by human cellular studies and that the non-human vertebrate studies in those reviews show mixed results. Several reviews reported on changes in ionic channels through the cellular membrane under RF-EMF exposure. Others concluded that the evidence for RF-induced ionic signaling was weak. The reviews presented mixed conclusions on whether RF-EMF exposure can induce the expression of heat shock proteins (HSPs). Most of the reviews concluded that there is no effect or a very limited effect of RF-EMF exposure on the production of reactive oxygen species (ROS). Two reviews concluded that RF-EMF exposure can activate isolated neurons. Those reviews that studied effects on gene-expression in non-human vertebrate cells stated that there were not enough studies to come to conclusions.

Animal studies

Several reviews demonstrated dielectric heating of animals and increases of body core temperature. The thermoregulatory response to a whole-body RF-EMF exposure is not different from the response to alternative heating methods. This response includes changes in effects on metabolic heat production, heart rate, and blood pressure. Those reviews that considered genotoxicity of RF-EMF exposure determined using *in vivo* studies found contradictory results. There are certainly studies that demonstrated genotoxic effect of RF-EMF exposure *in vivo*, but some of those are also criticized in the review studies. Several reviews have focused on RF-EMF-induced (transient) changes in permeability of the BBB. Some reviews conclude that BBB permeability can be altered at high (localized) SAR levels. Other reviews conclude that the evidence for such effects is weak. One review explained those mixed reports in literature by stating that earlier studies found effects, but more recent studies of higher quality could not reproduce such effects. Two reviews discussed effects of RF-EMF exposure on EEG signals and electric activity in the brain. One review discussed brain function and structure and reports on mixed results on those outcomes. One review reported on effects of RF-EMF exposure on properties of neurotransmitters. Several reviews state that animals can hear pulsed RF-EMFs above a certain threshold, so-called microwave hearing. However, they also report that there is little evidence that telecommunication signals can induce this effect. A very limited number of studies investigated effects of RF-EMF exposure on the endocrine system and the majority of those studies did not find an effect. Most reviews that focused on effects on the cardiovascular system and RF-EMF exposure study those effects as part of a thermal response to dielectric heating. Those studies that did not show a thermoregulatory response did not show any effects on heart rate and blood pressure. Reviews that investigated RF-EMF exposure and effects on the immune system and hematology reported on transient effects of RF-EMF exposure that could be part of a thermoregulatory response. Only one review considered effects of RF-EMF exposure in

this frequency range on the skin, this review presented mixed results. Some reviews focused on ocular effects of RF-EMF exposure. They reported on the existence of such effects, but these might be thermal in nature. Reviews on behavioural effects of vertebrates under RF-EMF exposure have reported on behavioural responses to dielectric heating and on mixed results in behavioural responses regarding non-thermal exposure.

Environmental studies

Environmental studies on RF-EMF exposure and vertebrate behaviour focused mainly on animal nesting, reproduction, orientation, and abundance near RF-EMF sources. There are a couple of reviews that concluded that behavioural effects might occur for birds and bats under RF-EMF exposure. Two studies on cows were reviewed that also showed effects of RF-EMF exposure during development. A few review studies reported on reproductive effects in birds exposed to environmental RF-EMFs. One review discussed effects of low-frequency RF-EMFs on birds' orientation.

5.1.2. Invertebrates

RF-EMF exposure of invertebrates in the 0.4-6 GHz frequency range has been studied by several authors. Dielectric heating of invertebrates using RF-EMFs is demonstrated in many studies and the dielectric properties of invertebrates in this frequency range have been studied as well. Most studies that do not aim to induce dielectric heating focus on developmental, genetic, or behavioural effects. In vitro studies of exposure of invertebrates' neural cells to RF-EMFs has shown to lead to increased neural activity. In vivo studies in laboratory conditions are faced with several problems and present inconclusive results on a series of investigated parameters. Studies with better exposure assessment of both the exposed groups, the sham-exposed groups, and the controls are necessary. Environmental studies present an interesting approach, in that sense that they, by design, use realistic exposure conditions. However, they are also faced with their limitations in terms of exposure assessment. Studies on non-insect invertebrates are underrepresented in this category (9/70 studies reviewed). Given the fact that all of the studies found effects of RF-EMF exposure (given the experimental shortcomings of some of those studies), it seems warranted to execute more research in that domain.

5.1.3. Plants and fungi

Dielectric heating of plants and seeds using RF-EMFs below 6 GHz is possible using high levels of RF-EMFs. This heating might have beneficial effects for some plants at very short exposure times, but will induce plant mortality after a certain exposure time. At lower levels of RF-EMF exposure, those effects that are demonstrated in literature seem to happen on a relatively short time scale and seem to occur for particular frequencies, modulations, or exposure durations. No studies were found that reproduced such effects. Studies on longer term exposure to low-intensity (in comparison to those RF-EMF levels necessary for dielectric heating) seem to show no effect, but the number of studies and studied plants and especially fungi is limited. Some interesting environmental studies have been proposed, but currently lack proper control. Future research in this area should focus on: (1) higher quality control and sham control groups, (2) monitoring temperature during the entire experiment, and (3) quantify the RF-EMF exposure of both the control and exposed groups over time during the entire experiment.

5.2. Higher telecommunication frequencies (6-300 GHz)

5.2.1. Vertebrates

Cellular studies

Several cellular studies demonstrated dielectric heating of cells. A limited number of studies on genotoxicity, with poor control and exposure assessment exist. Neural activation using pulsed RF-EMF was investigated using studies with high-quality control. Changes in parameters of the compound action potential under RF-EMF exposure were demonstrated. Studies on changes in cellular transformation showed no non-thermal effects. Other in vitro studies showed an increased ROS production in mouse neutrophils under RF-EMF exposure. It is unclear whether this effect is thermally induced or not. Some effects on ionic channel parameters were demonstrated, but were shown to be thermal in nature. No effect was found on cellular metabolism and membrane receptors of rats.

Animal studies

Several studies demonstrated (body-core) temperature increases due to RF-EMF exposure. At very high-power densities, this can lead to death of the vertebrates due to circulatory failure. Thresholds in terms of power density and exposure times have been determined for rats and mice and the behaviour of several body parameters (blood pressure, heart rate, core and skin temperature) during RF-EMF heating have been investigated. Behavioural aspects were investigated for animals exposed to much lower power densities. Mixed results on behaviour of animals in front of X-band radar were shown. Some studies showed changes in behaviour, some did not show an effect. Reproductive effects were investigated as well. Studies on animal sperm at 10 GHz showed reductions in sperm count for 52 days of exposure at a relatively high level. The effect seemed to be thermal. Mixed effects are demonstrated on the growth of injected tumor cells in rodents. Those studies that found an effect showed a reduction in tumor development. RF-EMF exposure of the eye can induce corneal lesions and cataract. However, there is a debate on what the actual threshold values are for the effect to occur. Some effects are described on neurostimulation in vivo, but the amount of studies is very limited. Several studies (from the same research group) demonstrated that RF-EMF exposure can have a hypoalgesic effect in mice. The effect of RF-EMF on immune responses was studied by several authors, most of those showed that high-frequency RF-EMFs can be used to induce an anti-inflammatory response, up to a certain dosage. Finally, one study found effects of RF-EMF exposure on EEG spectra.

5.2.2. Invertebrates

Dielectric heating of invertebrates in the 6-300 GHz frequency range was demonstrated in several references. Studies that investigate exposure to relatively high intensity RF EMFs have found effects on neural responses (in vitro) and on the development of insects (in vivo). Two papers presented experiments at RF-EMF levels below the ICNIRP basic restrictions in this frequency range and found some effects in insect development. More research at these exposure levels is needed to verify some of the demonstrated effects at realistic exposure levels. The number of in vivo studies on RF-EMF exposure of invertebrates in the higher frequency range is very limited and should be extended in the future.

5.2.3. Plants and fungi

Dielectric heating was demonstrated in the 6-300 GHz frequency range for plants. In order to demonstrate other effects, future studies should focus on proper exposure assessment of the exposed, control, and sham groups. Moreover, it should be studied whether sham exposure is necessary in these studies. The series of papers shown in Table 20 demonstrate that proper sham exposure can change interpretation of the results in this field drastically.

6. Policy options

Based on the review presented in this document and the conclusions drawn from the review, four policy options are suggested.

6.1. Funding research on environmental exposure to RF-EMFs

The guidelines that form the basis for policy-making regarding RF-EMF exposure in most EU countries are those issued by ICNIRP (International Commission on Non-ionizing Radiation Protection (ICNIRP) 2020). While the work done by ICNIRP is valuable for policy-making, it has to be noted that the scope of the ICNIRP guidelines is limited to humans. These guidelines only consider literature on substantiated biological effects that are harmful to human health. The ICNIRP guidelines do not focus on prevention of undesired biological effects of RF-EMF exposure of animals, fungi or plants. To prevent environmental effects of exposure to RF-EMFs, policy-making and legislation should be based on scientific literature that focuses on RF-EMF exposure of non-human vertebrates, invertebrates, plants, fungi and other organisms. Hence, if policy-makers want to implement protective policy-making regarding non-human organisms, they should base their decisions on other sources within scientific literature that focus on these organisms. This is not a straightforward task, because, as this review shows, there are areas of research in this domain that have been underexplored.

A first problem is the disparity between the number of publications that focus on vertebrates versus the number of studies that focus on other species. At those frequencies where the current telecommunication networks predominantly operate (0.4- 6 GHz), there are hundreds of high-quality peer-reviewed studies that focus on effects of RF-EMF exposure of vertebrates and humans, see for example the amount of publications cited in (Vecchia 2009). The literature on invertebrates in the same frequency range is smaller (approximately 100 publications, see Section 3.1.2), with a vast majority of those papers focused on insects. Within that category, the amount of papers that focus on RF-EMF exposure of non-insect invertebrates is very limited (< 10 peer-reviewed papers). The amount of publications on plants and fungi in the frequency range below 6 GHz (approximately 100 papers, see Section 3.1.3) is also small in comparison to the literature on vertebrates. Additionally, many of the papers on invertebrates, plants and fungi are faced with experimental shortcomings.

A second issue is the relatively small amount of available peer-reviewed publications on RF-EMF exposure of non-human organisms in the 6-300 GHz frequency band (approximately 250 in total). This is relevant because 5G networks will also operate at frequencies between 6 GHz and 300 GHz. This amount of publications is relatively small in comparison to the amount of literature available between 0.4 - 6 GHz. In this frequency range, there exist similar differences between non-human vertebrates, invertebrates, plants and fungi as in the lower frequency range. There is a reasonable amount of studies that focus on non-human vertebrates (< 150 publications). However, the peer-reviewed literature on invertebrates (<50 studies), fungi (< 15 studies) and plants (< 15 studies) is very limited.

In order to counter these shortcomings in the current scientific understanding, a first policy option can be to fund research that results in more high-quality studies on plants, fungi and invertebrates at frequencies below 6 GHz, and to fund high-quality research on non-human vertebrates, plants, fungi and invertebrates at frequencies between 6 and 300 GHz. The results of these studies could form the basis for developing evidence-based policies regarding RF-EMF exposure of non-human organisms.

6.2. Systematic measurement and monitoring of exposure to environmental RF-EMFs

In order to assess whether precautionary measures need to be taken in order to protect an organism from an exposure, two components are required. First, it needs to be proven that the exposure has a negative effect or there needs to be uncertainty on the effects of the exposure. Second, there has to be a risk for a(n) (significant) exposure to occur. Given the relatively small amount of published papers on RF-EMF exposure in some of the categories studied in this document (see Section 6.1), there is uncertainty on the effects of a potential exposure. However, the question remains what the exposure of non-human organisms to RF-EMFs will be.

As outlined in Sections 1.3-1.5 of this document, nearly all non-human organisms will fall into the non-user category in terms of RF-EMF. Hence, the dominant sources of RF-EMF exposure are far-field sources, so-called environmental exposure. In Section 1.5, it was shown that there are reasons to believe that this exposure is expected to change in 5G networks. However, since there are almost no 5G networks operational at the moment, it is difficult to predict this exposure. Therefore, a second policy option could be to call for or require systematic measurements or monitoring of environmental RF-EMFs.

Particular attention should be paid to those environments where non-human organisms are more prevalent, since most previous studies focusing on exposure to environmental RF-EMFs have a human-centric approach, where the vast majority of measurements take place in environments where the prevalence of non-human organisms is relatively low (Bhatt et al. 2016; Bolte and Eikelboom 2012; P. Frei et al. 2009; Sagar et al. 2016; 2018; Thielens, Van den Bossche, et al. 2018; Urbinello, Huss, et al. 2014; Velghe et al. 2019b). There are some environmental studies presented in this review, which focus on environmental exposure to RF-EMF of non-human organisms (Vijver et al. 2014; Lázaro et al. 2016; Mittler 1977; Pramod and Yogesh 2014; Balodis et al. 1996; M. Cammaerts and Johansson 2015; Haggerty 2010; Magone 1996; Waldmann-Selsam et al. 2016). This line of studies needs to be expanded.

The measurement protocols for measurements of RF-EMF exposure in 5G networks are currently being developed (Aerts et al. 2019) and can be used to measure environmental exposure to RF-EMFs. However, such measurements require a trained technician or scientist for execution and are time-intensive. An alternative would be to deploy RF-EMF monitoring networks (Aerts et al. 2018; Vermeeren et al. 2019; Dürrenberger et al. 2014). These are networks of nodes with the ability to measure RF-EMF levels and that are deployed strategically over an area in which the RF-EMF exposure should be monitored. Such monitoring networks have the advantage that they only have to be deployed once and provide temporal information without the need for a technician to go on site. There is of course a cost associated with the deployment of such measurement nodes.

6.3. Monitoring of base station antennas

An alternative to executing measurements of environmental RF-EMF exposure is to monitor the output power of the dominant source of environmental RF-EMFs: the base station antennas. Network operators regulate this output power, depending on the load in the network and the requirements of the users. It has been shown in literature (Shikhantsov et al. 2020) that, given the correct information on the used precoding on the base station antennas and the configuration on the antennas, it is possible to determine the environmental exposure caused by such base station antennas. This can be used on a larger scale in combination with the methods provided in (Beekhuizen et al. 2013; 2014; Bürgi et al. 2010). However, such information is not publicly available and telecom operators keep this information to themselves.

Therefore, a third policy option can be a request by policy-makers to make this information public, i.e. it can be requested that operators have to disclose their used antennas, operation frequencies, precoding used over time, output power over time, and specifications of the antenna installation. Alternatively, it is possible to install an independent expert committee that can interpret this data if there would be reasons (trade secrets etc.) for not disclosing this information publicly. This data can then be used as an input to the methods listed above to retroactively assess the RF-EMF exposure over time. Such information could be useful if new scientific insights should arise and it would simultaneously allow the operators to continue with necessary updates of their networks.

6.4. Compliance assessments and prevention of high RF-EMF exposures near base station antennas for all living organisms

There are situations where it is clear that a high RF-EMF exposure will occur: mobile animals can find themselves in very close proximity to a base station antenna or such transmitters can be installed in the vicinity of trees. In such cases it is possible to apply measures that will ensure physical separation between base stations and the exposed organisms that are similar to those currently applied for humans. The installation of such antennas is regulated and a compliance assessment based on the ICNIRP's guidelines is usually required. These guidelines contain relationships between basic restrictions on the specific absorption ratio (SAR), i.e. a proxy for thermal heating due to RF-EMF exposure, and the incident RF-EMF levels, the so-called reference levels. These basic restrictions and reference levels are commonly used to assess compliance of newly installed base station antennas (Thors et al. 2017; Baracca et al. 2018; Thielens et al. 2013), and they result in limitations on the allowed output power of these antennas and physical barriers that are placed around such antennas to prevent the general public from approaching them. Similar barriers could be installed to prevent airborne animals from appearing in close proximity to base station antennas. A minimal separation distance to existing plants can be determined based on measurements and numerical simulations.

A fourth policy option can be to require compliance studies for other organisms than humans when base station antennas are installed. These are studies that quantify the exposure of a subject near an antenna and result in a maximal output power and minimal separation distance for such antennas, based on the potential exposure and effect of such exposure that might occur. Since dielectric heating has been demonstrated in all categories studied in this review, this effect should be prevented for all organisms. These compliance studies should be executed for all organisms that are likely to appear near such an antenna and the emitted power of these antennas has to comply with the results of such studies. Typical examples here are bats, birds, insects and nearby plants.

The current compliance studies that are executed with focus on humans are not sufficient to prevent thermal effects in non-human organisms. The physical mechanism for heating due to RF-EMF exposure is the same in all biological materials. However, the relationships between RF-EMF exposure, dosimetric quantities and temperature elevations that are used in the ICNIRP guidelines are based on properties of humans and on experiments conducted using animals (predominantly vertebrates). These relationships are different for other organisms, which can have significantly different characteristics, such as: surface-area-to-volume ratios, dielectric properties, thermal properties, thermoregulation and physical size.

The main difference between the first suggested policy option and the fourth one is that the first one is focused on establishing more scientific insight in biological effects of RF-EMF exposure, while the one suggested in this section calls for technical improvements of the compliance of base station antennas. The demonstration of prevention of dielectric heating in other organisms than humans is possible with the currently existing scientific methods.

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Telecommunication networks use radio-frequency electromagnetic fields to enable wireless communication. These networks have evolved over time, and have been launched in successive generations. The fifth generation of telecommunication networks will operate at frequencies that were not commonly used in previous generations, changing the exposure of wildlife to these waves. This report reviews the literature on the exposure of vertebrates, invertebrates and plants to radio-frequency electromagnetic fields in anticipation of this change.

The review shows that dielectric heating can occur at all considered frequencies (0.4-300 GHz) and for all studied organisms. Summarising and discussing the results of a series of studies of radio-frequency electromagnetic field exposure of wildlife, the review shows that several studies into the effects of radio-frequency electromagnetic field exposure on invertebrates and plants in the frequency bands considered demonstrate experimental shortcomings. Furthermore, the literature on invertebrate and plant exposure to radio-frequency electromagnetic fields above 6 GHz is very limited. More research is needed in this field.

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