by Hydroxyl Technologies

The Testing & Verification of Airora's Technology

1 Introduction

The hydroxyl radical cascade created by Airora's technology has been tested in world class laboratories and proven effective at inactivating both air and surface borne pathogenic viruses and bacteria with an up to 6 log (99.9999%) kill rate.

Hydroxyl Technologies Ltd, owner of the Airora brand, was born from the technological development previously achieved by Tri-Air Developments Ltd (2006 – 2017). Tri-Air took the novel hydroxyl cascade technology created by inventor Alan Mole and in partnership with BRE Ltd undertook over 10 years of prototyping and extensive microbiological and air quality testing at world leading laboratories, including the UK Health Protection Agency (HPA) at Porton Down.

Tri-Air underwent a change of investors in late 2015, which led to some of the original founding directors, led by Lorraine Baldry and Alan Mole, acquiring all of the Tri-Air intellectual property rights and forming Hydroxyl Technologies Ltd (HTL) to continue development, the latest outcomes of which can be found at airora.com.

The journey has involved the expert and professional capabilities of several internationally renowned independent design, research and testing organisations, including:

- The UK Building Research Establishment's (BRE) Internal Air Quality Team and Laboratories
- The UK Government's Health Protection Agency (HPA) at Porton Down
- The UK Government's Health and Safety Executive (HSE) Laboratory
- The University of Leeds
- The University of York
- The University of Ottawa
- PA Consulting

1.1 Hydroxyl radicals

Discovered by the UK's Ministry of Defence in the early 1960s, hydroxyl radicals (originally called the 'Open Air Factor', often just called 'hydroxyls') are highly reactive molecules of oxygen (O) and hydrogen (H); their chemical formula is OH.

Hydroxyls are continually produced in abundance in the troposphere (where we live) and wage a constant war of attrition against contaminants such as allergens, pollution, viruses, and bacteria.

When hydroxyls are created, they immediately seek out and react with contaminants in the air and on surfaces. These reactions happen within seconds and break down larger molecules, such as volatile organic compounds (VOCs), and tiny structures such as viruses and bacteria. Hydroxyls are so effective at cleaning and sanitising the air that scientists call them ***"Nature's Detergent"*** - a term first coined by Nobel Prize winner Paul Crutzen.

Proprietary and third-party data show that hydroxyl radicals:

- kill (inactivate) all types of both air and surface borne pathogenic viruses and bacteria that are known to harm people, including those in the Coronavirus family, Colds, Flu, MRSA, C-difficile and Norovirus
- neutralise airborne allergens such as pollens, spores, pet dander, cat saliva and house dust mite excretions
- eliminate common lung irritants and other pollutants such as ozone, volatile organic compounds (VOCs), formaldehyde and carbon monoxide
- remove malodours from the air

There are, on average, more than two million hydroxyls in each cubic centimetre of outdoor air during daylight hours.

1.2 How Airora works

Airora's technology mimics the effect of the natural chemistry of open air to create a safe and natural hydroxyl radical cascade throughout every corner of a room.

Inside the device the technology initiates the production of hydroxyl radicals, which are emitted alongside trace levels of ozone and linalool (a terpene). The linalool preferentially reacts with and destroys the ozone in a cascade reaction which creates an abundance of hydroxyl radicals.

The University of Leeds, one of the few leading laboratories in the world which is able to measure hydroxyl radical density, has confirmed that our technology creates hydroxyls at a rate which aligns with typical ambient conditions at noon in the summertime.

Our technology is designed to operate 24/7 and has been shown to continuously suppress high levels of pathogens, both in the air and on surfaces.

2 Efficacy

2.1 The inactivation of pathogenic viruses and bacteria

Atmospheric hydroxyl radicals are proven to kill human pathogens, be they bacteria or viruses, by a well understood process: In general terms, hydroxyls react with the lipids and proteins in their thin, delicate surface structures, causing lysing (breakdown).

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Our extensive test data is from several different prototypes over 12 years and includes both laboratory tests and field trials:

Test Facility / Date	Prototype Unit Tested	Test Type	Test Microbes	Test Method	Test Chamber Size	Results
HPA UK Jun 2006	Proof of Concept unit	Airborne	MS-2 Coliphage	Aerosolisation (high concentrations)	18m3	6 log (99.9999%) kill in less than 5 minutes
HPA UK Sep 2007	Plasmalyser Prototype no.1	Airborne	1) Bacillus atrophaeus (gram +) "aerostable spore" 2) Staphylococcus epidermidis (gram -)	Aerosolisation (high concentrations)	18m3	1) 1 to 2 log (99%) kill in 60 minutes 2) 5 log (99.999%) kill in 2 minutes
HPA UK Oct 2007	Plasmalyser Prototype no.1	Surface (steel)	1) Bacillus atrophaeus (gram +) 2) Staphylococcus epidermidis (gram +3) MRSA	Tested at 2.5, 3, 3.5 and 4 hrs.	18m3	1 & 2) 0.5-1 log kill in 4hrs 3) 1 log kill in 4 hrs and a 5 log kill in 2.5hrs
HPA UK Nov 2007	Plasmalyser Prototype no.2	Surfaces (steel & glass)	1) MRSA (low concentrations) 2) MRSA (high concentrations)	1) Tested over 1 & 4hrs. 2a) Tested at 24 hrs & 2b) Tested at 48hrs.	18m3	1) At 1 hour NO surviving MRSA on glass or steel. 2a) NO surviving MRSA on glass (greater than 6 log kill) & a 3 log kill on steel 2b) 4 log kill on steel
FDA lab USA Sep 2008	Plasmalyser Prototype no.2	1) Airborne 2) Surface (steel / formica / textile)	Test 1) Bacillus atrophaeus Test 2a) MRSA & 2b) Bacillus atrophaeus (high concentrations)	1) Aerosolisation (high concentrations) 2) Tested over 26 – 36hrs.	15m3	1) 6 log kill after 20 minutes 2a) 3 log kill on all surfaces at 14-16hrs 2b) 2 log kill on steel & formica & 4 log kill on textile at 14-16hrs.
HPA UK Feb 2010	T250 prototype (unit 1 & 4)	Airborne	MS-2 Coliphage	Aerosolisation (high concentrations)	0.9m3 safety cabinet	Unit 1 achieved 5.8 log kill in 30 mins. Unit 4 achieved 4 log kill in 1-2 hours.
HPA UK Feb 2010	T250 prototype (unit 4)	Surface (stainless steel)	MS-2 Coliphage & MRSA	Both tested at 0, 2, 4, 24 & 48hrs.	0.9m3 safety cabinet	1.5 log reduction at 24 & 48 hrs
Real world Coffee Shop 1 week in Jul 2010	T250 product	Airborne	1) Bacteria 2) Fungal Spores 3) Enterobacteriaceae	Real world samples taken from a working coffee shop over 1 week.	Approx 300m3 with front & rear entrances in constant use.	1) 95% kill 2) 90% kill 3) 100% kill
Real world Cinema 1 week in Jul 2010	T250 product (2 units)	Airborne	1) Bacteria (human) 2) Bacteria (environmental)	Real world samples taken from a working cinema auditorium over 1 week.	Approx >1000m3	1) 95% kill 2) 86% kill Cinema staff positive on effect on malodours
Real world Care Home 1 week in Jul 2010	T250 product	Airborne	1) Bacteria (human) 2) Bacteria (environmental)	Real world samples taken from a working care home which had the windows open during the day.	Approx 400m3 sited in main dining hall	1) 91% kill in dining hall & 98% kill in common room 2) 90% kill in dining hall & 89% kill in common room

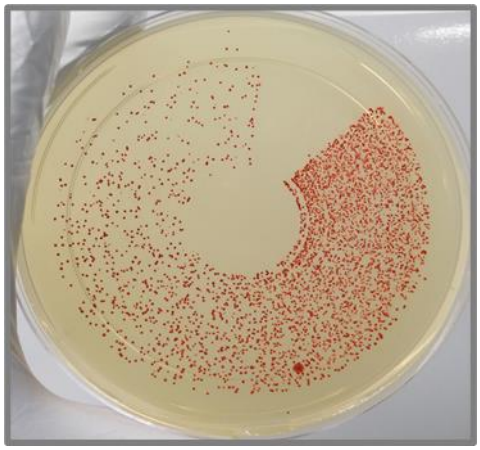
2.2 Testing by Ottawa University in 2013

In 2013 Ottawa University undertook testing on the T60 prototype on behalf of an interested third party. A summary of the results is shown below:

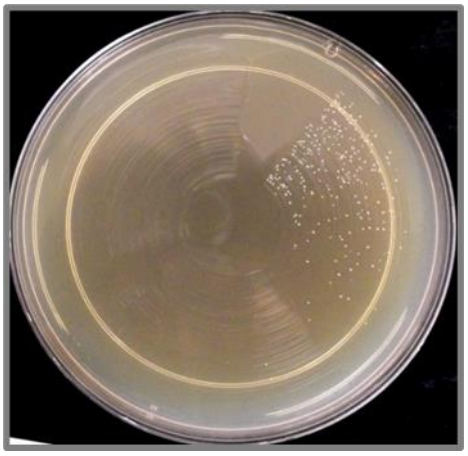
Photographs of recovery plates from the 120-minute Reyniers slit air sampler:

The first stained photograph of the control plate (SE #1) shows the colony forming units (CFU) of Staphylococcus epidermidis recovered from the air of the aerosol chamber. The second photograph represents a preliminary examination (20 hours of incubation) of unstained bacteria of the test plate (T60- #1-1) representing the first evaluation of the development prototype (LED/35ppb O3).

Control Plate: Stability in Air (SE #1)



Test Plate: Device Test (#1-1)



It can be seen that the S. epidermidis has been reduced in numbers to undetectable in ~35-40 minutes (the challenge input – nebulizer fluid concentration of the SE#1 was 4.9Log10 and the #1-1 was 4.82Log10) these values show that the inoculums prayed into the chamber are consistent and these plates can be compared to each other for the purposes of efficacy.

In addition, the University of Ottawa also undertook testing on the T250 prototype, with the following results:

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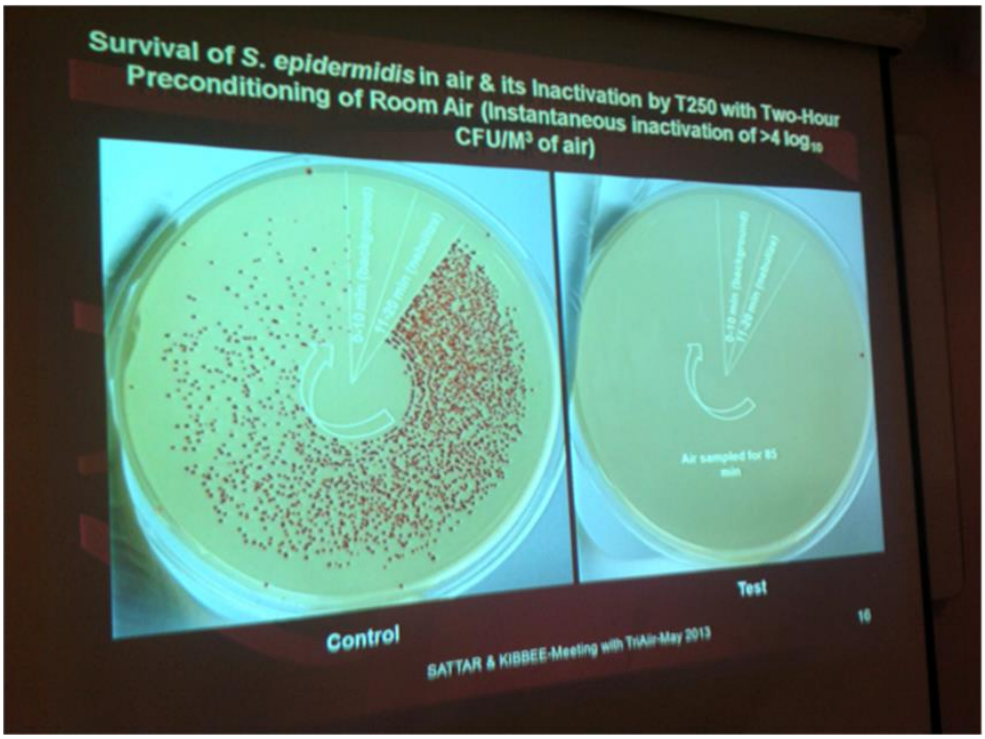
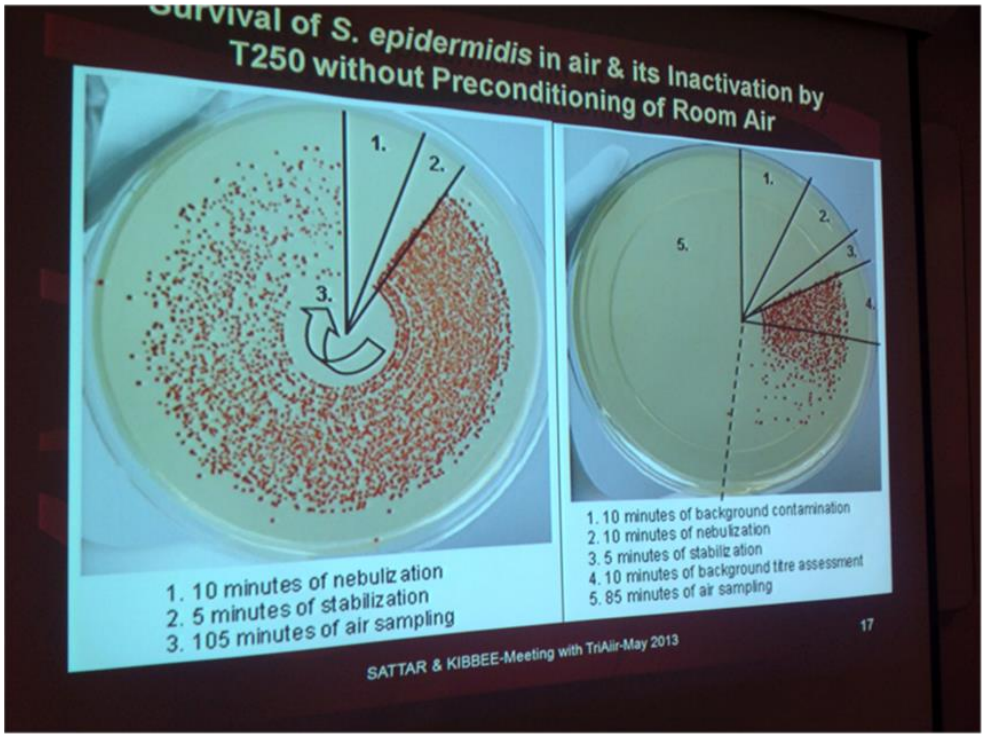
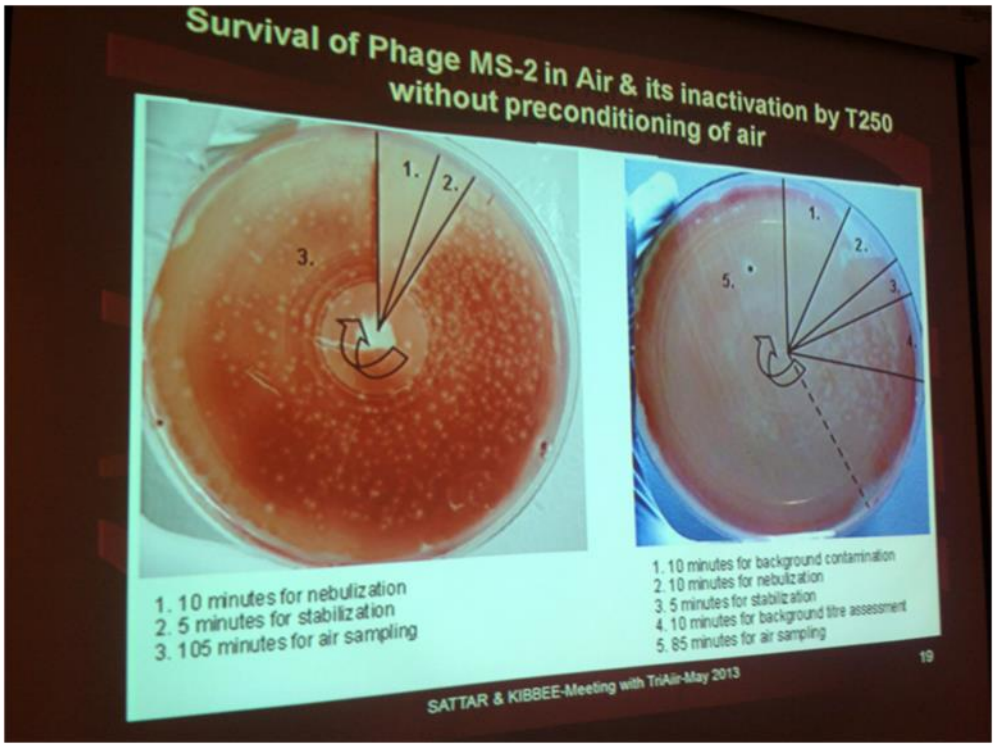
Survival of S.epidermidis in air and its inactivation

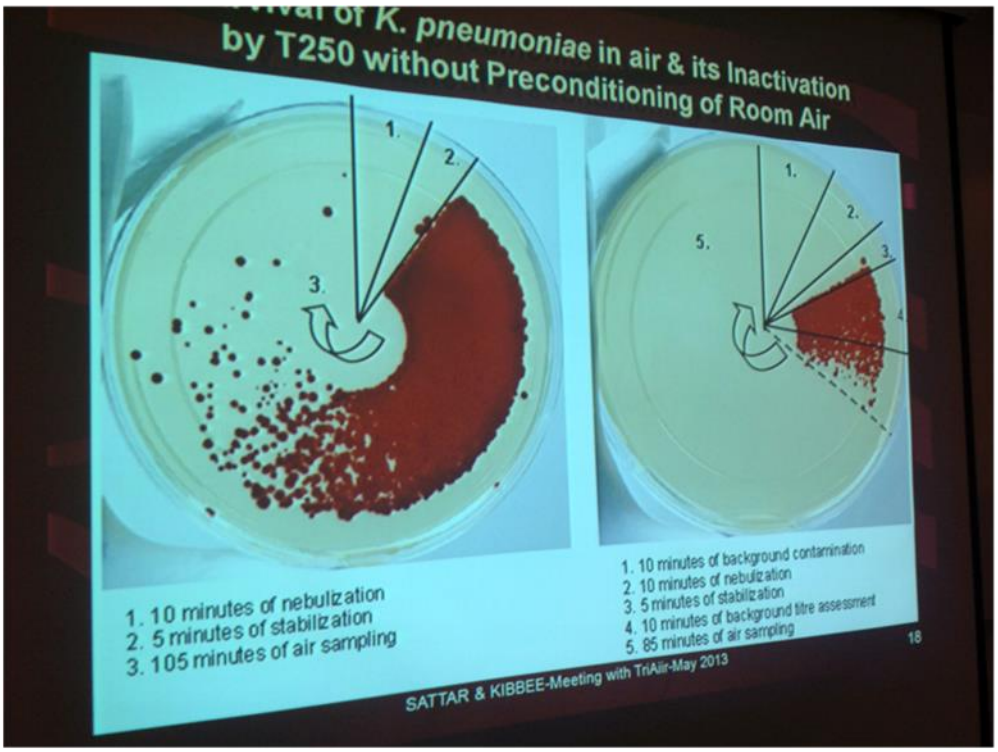




Survival of Phage MS-2 in air and its inactivation



Survival of K. pneumoniae in air and its inactivation



2.3 Removal of VOCs and other pollutants / asthma triggers

It is well documented that the powerful oxidisation effect of hydroxyl radicals eliminates pollutants and common lung irritants such as ozone, volatile organic compounds (VOCs), formaldehyde and carbon monoxide.

BRE undertook extensive air quality testing of various Airora prototype devices. As part of these tests, BRE attempted to qualify and quantify the test chamber air prior to, during and after testing, to ascertain exactly the air emissions created by the Airora technology versus the background air.

BRE has carried out extensive testing of an Airora development prototype to measure its performance in reducing VOC concentrations in the air.

The following VOCs, typical of domestic life, were employed in the tests:

VOC	CAS Number	Boiling Point °C	Reason for Inclusion
Butan-1-ol	71-36-3	117	Representative of the family of alcohols, including ethanol (alcohol, perfume), propan-2-ol (sanitising gel, cleaning agent, IPA).
Butan-2-one (Methyl ethyl ketone, MEK)	78-93-3	80	Representative of ketones, a common solvent in industrial paints, glues.
Dodecane	112-40-3	215	Representative of alkanes, aliphatic hydrocarbons often found in paints, glues, fuels
Limonene	138-86-3	179	Representative of terpenes, found in air fresheners, scents, as well as citrus fruits.
Octamethyltrisiloxane (OMTS)	107-51-7	153	Representative of siloxanes, often found in personal care products and cosmetics, as well as dry cleaning.
Toluene	108-88-3	111	Representative of aromatic hydrocarbon compounds found in pyrolysis products and solvents.

Identical VOC concentrations were released into a controlled environment, both where no Airora device was active and where an Airora device was active and the results are shown below:

2.4 VOC decay with a prototype Airora device

VOC	RT	Time in minutes after spiking Concentration in air								
		5	30	60	90	120	150	180	210	240
C4 hydrocarbon	5.05	2385	1708	1243	966	785	233	434	344	325
C5 hydrocarbon	6.20	8	7	6	6	6	8	6	6	6
Butan-2-one	8.00	194	126	88	61	41	29	21	17	16
Butan-1-ol	10.95	134	185	125	83	57	39	31	26	25
X2	11.70	4	3	4	4	3	4	3	3	3
Toluene	13.15	183	168	120	79	54	36	27	22	19
Octamethyltrisiloxane	16.75	28	58	71	55	38	23	18	14	13
Limonene	27.35	26	49	80	68	50	29	18	14	10
Linalool	33.05	5	3	5	4	4	3	4	5	3
Dodecane	33.60	7	13	30	34	36	36	39	30	40
TVOC		**473**	**484**	**449**	**342**	**255**	**193**	**146**	**132**	**118**

- All concentrations are in μgm^{-3}.
- RT denotes the approximate retention time in minutes, rounded to the nearest 0.05 minute.
- ND denotes less than 2 μgm^{-3}.
- C4 and C5 hydrocarbons are not accurately quantified by this technique.

2.5 Formaldehyde

In nature, formaldehyde is produced from a variety of sources. As hydroxyl radicals decompose larger VOCs, smaller and smaller hydrocarbon compounds are formed. Formaldehyde is the smallest; it contains one carbon bonded to oxygen (H2C=O) and represents the last step before the final oxidation to produce carbon dioxide – carbon bonded to two oxygen atoms (O=C=O).

Formaldehyde is a common contaminant in indoor air, as it outgases from fabricated wood products, paints, adhesives, fabrics etc. It is toxic and the US Occupational Safety & Health Administration (OSHA) guideline for formaldehyde exposure is less than 750 ppb (0.75 ppm) over an 8-hour period. Short-term exposure cannot exceed 2 ppm. Formaldehyde reacts rapidly with hydroxyl radicals, so hydroxyl radical generators used indoors are very effective in decomposing it.

The testing facilities at BRE filter out as many contaminants as possible from the air entering the test chamber prior to an Airora emissions test. It is impossible to remove

all background compounds and some formaldehyde was inevitably present (formaldehyde is emitted by many surfaces used in the construction of the built environment). Throughout the many tests undertaken by BRE for HTL during the development of the Airora prototypes, it has been shown that the technology breaks down and removes formaldehyde over time.

The data collected from the final low, medium and high device settings for the T60 development prototype illustrate this effect:

Test/Setting	Formaldehyde µg/m^3		
	Before test	At end of test	Change
1 Low	4	5	+1
2 High	7	6	-1
3 Medium	6	6	0
4 Low	10	2	-8
5 Medium	7	5	-2

References:

1. B. J. Finlayson-Pitts and J. N. Pitts, Jr., The Chemistry of the Upper and Lower Atmosphere, Academic Press, San Diego, 1999.
2. J. A. Logan, M. J. Prather, S. C. Wofsy, and M. B. McElroy, Atmospheric Chemistry: Response to Human Influence, Phil. Trans. Roy. Soc. (London) 290, 187 (1978).
3. C. J. Weschler and H. C. Shields, Production of the Hydroxyl Radical in Indoor Air, Environ. Sci. Tech. 30, 3250 (1996).

2.6 Airborne Allergens

Independent third-party research relating to hydroxyl radicals and their effect on indoor allergens illustrates how hydroxyl radicals can prevent the onset of allergic reactions.

Pollens, Fungal Spores, Pet Dander

Hydroxyl radicals have been shown to reduce IgE-binding capacity in pollens, spores and pet dander through the degradation and modification of the tertiary structure and/or the induction of protein denaturation and/or aggregation. This allergen structure is then no longer recognised by the body's immune system and therefore histamine and other chemical mediators are not released.

While the references below refer in their titles to cluster ions, the text makes it clear that the recorded effects are achieved by hydroxyl radicals which result from the chemical interactions between the cluster ions.

References:

1. Kawamoto S et al. Decrease in the Allergenicity of Japanese Cedar Pollen Allergen by Treatment with Positive and Negative Cluster Ions, International Archive of Allergy and Immunology, 2006, Vol.141, No. 4
2. Kazuo Nishikawa et al. Exposure to positively and negatively charged plasma cluster ions impairs IgE binding capacity of indoor cat and fungal allergens, World Allergy Organization Journal 2016

House Dust Mites

Hydroxyls instantly denature the allergen Der p1 and Der f1 found in house dust.

Hydroxyls oxidise their protein structures, for example protein backbone damage due primarily to a hydrogen atom abstraction at the alpha carbon. This process leads to backbone fragmentation.

Side-chain damage is another possible protein oxidation mechanism and can occur through hydrogen abstraction or oxygen addition. Both hydroxyl radical initiated oxidation mechanisms result in a modified allergen structure. This allergen structure is then no longer recognised by the body's immune system and therefore histamine and other chemical mediators are not released.

References:

1. Garrison W M. Reaction mechanisms in the radiolysis of peptides, polypeptides, and proteins. Chem Rev 1987:381-398 -9920.
2. Singh J & Thornton J M. Atlas of Protein Side-Chain Interactions, Vols. I & II, 1992 IRL press, Oxford.

2.7 Malodours

Malodour experiments are undertaken either by chemical analysis or by human noses. While there is extensive literature demonstrating the ability of hydroxyl radicals to react with, break down and remove malodours, HTL has also undertaken field tests to demonstrate the effects of Airora on malodours.

Blindfold Test – Consumer Reactions July 2013

Airora has undertaken several human nose experiments, the most significant of which was in 2013. Using a professional consumer research firm, HTL took 12 groups of recruited consumers, both women and men, through a blindfold experience of an Airora device inside two different test houses. Each of the groups was then de-briefed in sessions which were captured on video (over 7hrs), with a two-minute video summary (available under NDA) produced to illustrate the overwhelming reactions of the groups.

The language used by these consumer groups to express the beneficial effect of the T60 prototype included:

- “outdoors”
- “in a spa”
- “on holiday by the sea”
- “feel energised”
- “motivated”
- “mountains”
- “forests”

3 Safety

3.1 Air Quality and Trace Compounds

Throughout the development of the technology the utmost concern has been to ensure the safety of the devices. To this end there has been extensive testing of the emissions starting with the earliest prototypes.

The Indoor Air Quality team at BRE undertook most of this work, with additional input from the University of Leeds and University of York.

At the outset a thorough study was carried out by BRE to determine international regulations and limits for various relevant compounds. From this the tightest limits were taken as the starting point for compliance. However, working with various third parties, who were interested in the technology, key limits were further tightened.

The testing at BRE was carried out in a specialist 18m^3 chamber designed for air quality studies. This was chosen as it represented a small bedroom sized space reflecting the smallest room in a house where a consumer version of the device might be used. Testing in a small chamber like this represents a tougher challenge from an air quality point of view.

The results below are for a recent prototype, which could be set to low, medium, and high outputs:

Parameter	Limit	Units	Low Setting	Medium Setting	High Setting
Ozone	50	ppb	6	11	30
TVOC	100	µg/m^3	5	7	8
Formaldehyde *	2.5	µg/m^3	-8	-4	-1
PM2.5	15	µg/m^3	<2	<2	<2

*The negative formaldehyde values are because the device had reduced the original background level of the test chamber.

BRE and the University of York were, together, able to identify all trace compounds resulting from the use of our technology and were able to confirm that none are known to be hazardous.

3.2 Linalool

Airora's technology aerosolises a trace amount of linalool at the final stage of the device, which is emitted into the room. Linalool is a terpene, and is the final component required in the formation of a hydroxyl cascade throughout the room.

Linalool has been assessed as safe by a multi-national body. See report here.

Technology Development Timeline

2007	Extensive microbiological testing of prototype engines at the UK Government's Health Protection Agency at Porton Down.
	Granted initial patent in GB followed by 60 jurisdictions.
2008	Prototype designated T250 developed leading to radical steps in the understanding of the technology and device design.
2010	T250 learning leads to concept design of 'domestic' prototype room scale devices, termed T35 (without a fan) and T50 (with a fan).
2011 - 2013	In-depth independent testing to confirm all aspects of safety and emissions and the associated regulatory requirements worldwide, with a target to reduce all emissions to < 1/10 of the acceptable regulatory limits.
2013	Prototype T60 developed to embody and exceed all regulatory and other requirements.
	T60 subject to further extensive efficacy and safety testing.
2014	Additional, new patents applied for and subsequently granted.
2015 – 2016	Further developments and simplifications with a focus on allergens and asthma, leading to the development of the Airora Home.
2017 - 2019	Final industrial design and testing of Airora Home. Development of a personal wearable device, Airora Personal.
March 2020	Collaboration with PA Consulting to rapidly develop and bring to market the Airora Professional, a wall-mounted product designed for all types of commercial and public buildings, including Hospitals, Care Homes, Schools, Offices and Airports.