Science Digest

Michael Legge

Sleep and creeping tumour cells

Despite the new technologies and therapies cancer still remains a leading killer and constantly reveals gaps in our knowledge both about the many types of cancer and the metastatic spread. Metastasis is linked in up to 90% of all cancer deaths and has been linked to circulating tumour cells (CTC) believed to be cells "broken off" from the original tumour. These cells retain their ability to proliferate and can associate themselves with immune cells thereby increasing their aggressiveness. Such dynamics is poorly understood. However, a recent publication (1) indicates that the release of CTC is connected to the sleep cycle. It is well known that the circadian rhythm can influence cancer development and the authors of the recent publication now provide evidence for this. Using 30 breast cancer patients as a "model' the authors took blood samples at 4am and 10am, representing "resting" and "active" times. The outcome was that 78% of all the cancer tumour cells identified were from the resting phase samples. They next used a mouse cancer model which demonstrated the same effect and when they disrupted the mouse sleep patterns the CTC still peaked during the resting phase. When CTC from the resting and active phase were injected into disease free mice, the resting phase CTC were more aggressive at forming tumours than the active phase mice. They then went on to identify a rise in human CTC cells gene expression associated with cell proliferation but the active CTC expressed an increase in protein synthesis. The authors conclude, that aggressive CTC occurs predominantly during sleep and may have far reaching implications for both treating cancer and the development of new cancer bio-markers.

Long COVID-19

While the world is gradually recovering from the COVID-19 pandemic, a new, phenomena is starting to emerge: long COVID. Three recent publications have drawn attention to long-COVID complications heart disease and COVID "brain fog". An early multi-centre study from the USA indicates that only 25% of people who were hospitalized directly for COVID-19 feel fully recovered after one-year and the team have identified immune markers that are associated with the worst cases (2). The study compared more than 150,000 people who recovered from COVID-19 with unaffected peers and a pre-pandemic group of people. For those admitted to intensive care, they had a higher, 20-fold risk, of cardiac swelling and blood clots compared to the unaffected peers. People who had not been hospitalized who had an associated increase risk of 8% in heart attack risks, and a 247% risk of cardiac inflammation. While the data is observational other smaller international studies support these findings. The original authors speculated that because the COVID-19 virus binds to the angiotensinogen converting enzyme-2 receptor (ACE-2), which is in many body cells including the heart it gives the virus access to the cells. This leads to endothelial cell damage causing the most likely formation of blood clots that have been consistent with COVID-19 infections. In addition, activation of the immune system may cause organ damage.

In two further publications (3,4), consideration is made to how COVID-19 "brain-fog" occurs as part of the complication of long-COVID as well as further long-term complications It has been well established that cognitive dysfunction ("brain-fog") is strongly associated with long-COVID. The authors identified that there were similarities between COVID-19 cognitive dysfunctions and another cognitive syndrome (chemobrain) which patients experience during chemotherapy and radiation therapy. Chemobrain is strongly associated with elevations of neurotoxic cytokines and reactive brain macrophages. Using mice as a model the current authors identified changes in the brain associated with COVID-19 and found that brain resident macrophages were elevated, and that CSF cytokines were also elevated. These effects persisted up to seven-weeks post-infection. Similar cytokine results were found in patients with COVID-19 and persisted during the long COVID-19 recovery period. However, only cytokine CCL-11 remained persistently elevated and the authors proposed that CCL-11 was responsible for modifying brain function by mounting neuro-inflammatory changes. It was further proposed that cytokine profiling may provide a better way of identifying and reducing COVID-19 "brain-fog".

Gender identity and Clinical Chemistry results

Transgender identity and the transition to transgender males and females is not uncommon. However, from a pathology laboratory perspective it can present issues in interpreting results. In the present publication (5) from the USA the authors have investigated the influence of transgender treatment on clinical chemistry reference ranges. Primarily, transitioning to a male involves testosterone therapy which provides the body changes associated with males. To transition to a female, estradiol is the main hormone therapy, however some transgender people may also use progesterone or androgen blockers. Pharmacologic therapies for both groups have the potential to change clinical chemistry indices such as creatinine, sodium, and potassium as well as haemoglobin and haematocrit values. In the present study 175 transgender people participated, comprising of 82 transgender men and 93 transgender women. Clinical chemistry samples were analysed on both Beckman and Roche platforms. Overall, 14 analytes were reported for both transgender groups, In general, the analyte pattern shifted towards the affirmed gender with their respective reference ranges and any changes between the two groups were consistent with their respective hormone therapy. The authors considered that gender-affirming hormone therapy had minimal impact on the majority of clinical chemistry results. However, they caution that the effects of the hormones on lipids was complex and that they did not control for diet and lifestyle.

Clinical utility of plasma from apheresis v^s whole blood

Fresh frozen plasma is indicated for a number of clinical conditions such as replacement of coagulation factors, and massive blood transfusions. While many studies have investigated different storage conditions on coagulation profiles, many lack data on the collection procedure. Plasma can be prepared from a single donor by plasmapheresis or by collecting from whole blood. In the publication from Egypt the authors evaluate the impact of collection procedures on the in-vitro quality of the plasma (6). Two plasmapheresis units were used, (Trima Acel, Terumo, BCT and Autopheresis-C, Freenius, Kabi Global) and whole blood collected from citrate-phosphate-dextrose-adenine (CPDA-1) blood. All plasma samples were kept at 4^{o}C for two days and coagulation parameters analysed on day 0 and day 2. The impact of freezing twice was also investigated. Immediately after collection three aliquots of each of the units was frozen at -20^{o}C. Only blood group A samples were used. Day 0 represented the first thaw and the second aliquot was stored at +4^{o}C for 24 to 48 hours, the third aliquot was refrozen. All samples were analysed for prothrombin/INR, aPTT, fibrinogen and Factors V and VIII. Whole blood plasma demonstrated a significant decrease in all coagulation parameters across the time periods except for fibrinogen. No significant differences were observed between either plasmapheresis units and both demonstrated greater factor activity. Re-frozen samples demonstrated a significant decline of all coagulation values. The authors concluded that coagulation factors were well preserved in the thawed specimens from either plasmapheresis system.

COVID-19 and ethnic minorities

As a result of a coordinated public health action in the UK, data has been gathered on the impacts of COVID-19 in the general population. In a report by Public Health England death rates for black, Asian and other minority groups has been shown to be higher than those in white British people.

In particular those with a Bangladeshi background were between 10% to 50% higher after accounting for age, sex and social status. In a recent publication from the UK a retrospective analysis of the biochemical data of patients admitted to hospital during the peak COVID-19 outbreak (April 1st to 28th 2020) was undertaken (7). All biochemistry data was obtained from Roche c-702 and 3801 analysers. A total of 311 patients were admitted (211 ethnic minority and 100 white British). While all groups had significantly elevated CRP the ethnic minority group was significantly higher than the white British group. Troponin-T was significantly higher in the white British group. AST and GGT were significantly higher in the ethnic minority group but ALT lower than in the white British group, however albumin and total bilirubin were not different between groups. Electrolytes and renal function parameters did not differ between all groups. The authors concluded that understanding the differences between the groups may contribute to a better understanding relating to why ethnic groups are at greater risk of death with COVID-19 and should be considered when interpreting biochemical data.

Safety first- three person embryos

About one in 5000 children are born with an inherited disease related to mutations in the mitochondrial DNA. Although only accounting for approximately one-percent of the total DNA in a cell, these mutations lead to multiple faulty organ function including the heart and brain and are ultimately fatal in early childhood. As mitochondria are completely inherited from our mothers (no paternal involvement) these serious disorders are only maternally inherited as at fertilization all the sperm mitochondria are destroyed when entering the egg leaving only maternal mitochondria. Mitochondria are essential for life providing approximately 98% of the energy requirements for normal metabolism via the complex respiratory chain. A mutation anywhere in the mitochondrial respiratory chain will result in a cellular failure to generate energy and all cells in the body with the exception of red cells contain mitochondria in vary amounts. There are seven well defined mitochondrial disorders and data from Australia indicates the approximately 20 children a week are born with these, largely untreatable disorders. Research from the UK has demonstrated that it is technically feasible to overcome these disorders using IVF and egg manipulation by either transferring the maternal chromosome spindle prior to fertilization or the maternal pro-nucleus formed shortly after fertilization to a second donor (unaffected) egg with the donors spindle or pro-nucleus removed. The resulting child would have the donor's disease-free mitochondria, thereby creating the so-call three parent embryo. The question arises about 'normal' development. A recent publication from China (8) using donated human eggs has for the first time indicated that transfer of the maternal spindle (maternal chromosomes only) to the donor egg does not influence early pre-implantation embryo development or the genetic make-up of the developing human embryo with a slight delay in the initiating of the embryo's methylation processes. This technique provides a potential treatment for the fatal mitochondrial disorders by using the egg donor's mitochondria. The use of this technology for human treatment has been approved in the UK and Australia but not in China or New Zealand.

REFERENCES

1. Diamantopoulou Z, Castro-Giner F, Schwab FD, Foerster C *et al*. The metastatic spread of breast cancer during sleep. *Nature 2022.* https://doi.org/10.1038/s41586-022-04875-y
2. Xie Y, Xu E, Bowe B, Al-Aly Z. *Nature Medicine.* Long-term cardiovascular outcomes of Covid-19. *Nat Med* 2022; 28(3): 583-590
3. Evans RA, McAuley H, Harrison EM et al. Physical, cognitive and mental health impacts of Covid-19 after hospitalisation (PHOSP-COVID): a UK multicentre, prospective cohort study. *Lancet Respir Med* 2021; 9(11): 1275-1287
4. Xu E, Xie Y, Al-Aly Z. Long-term neurologic outcomes of COVID-19. *Nat Med 2022;* https://doi.org/10.1038/s41519-022-02001-z.
5. Humble RM, Greene DN, Schmidt RL, McPherson GW. Reference intervals for clinical chemistry analytes for transgender men and women on stable hormone therapy. *J Appl Lab Med* 2022; 7: 1131-1144.
6. Hussein E, Enein AA. Quality assessment and clinical utility of plasma obtained via apheresis vs that obtained from whole blood. *Lab Med* 2022; 53(5): 439-445.
7. Biochemical abnormalities in COVID-19: a comparison of white versus ethnic minority populations in the UK. *J Clin Pathol* 2022;75: 663-667.
8. Qi S, Wang W, Xue X, Lu Z et al. Single cell multiomics analysis of spindle-transferred human embryos suggests a mostly normal embryonic development. *PLOS Biol* 2022; https://doi.org/1`0.1371/journal.pbio.3001741